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THE EFFECTS OF CRUDE OIL AND THE DISPERSANT COREXIT 9527 ON THE VEGETATION OF A NOVA SCOTIAN SALTMARSH: IMPACTS AFTER TWO GROWING SEASONS

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by

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THE EFFECTS OF CRUDE OIL AND THE DISPERSANT COREXIT 9527 ON THE VEGETATION OF A NOVA SCOTIAN SALTMARSH: IMPACTS AFTER TWO GROWING SEASONS

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ABSTRACT

This study documented the effects of experimental applications of Alberta sweet blend crude oil on the creek edge, midmarsh and high marsh zones of a Nova Scotia salt marsh over the course of two growing seasons. It also investigated the impact of the dispersant Corexit 9527 on the creek edge vegetation zone. The effects of the oil and dispersant applications were monitored using a number of parameters, including; plant height growth, stem density, biomass, species cover, plant fluorescence induction measurements, anatomical differences and soil profile changes. These data revealed that there was considerable variation in the sensitivity of the three vegetation zones to the oil treatment. The oil treatment had little effect on the creek edge zone (dominated by tall Spartina alterniflora) during the 1986 growing season and no residual effects were noted in 1987. The midmarsh zone (dominated by dwarf S. alterniflora) by contrast was severely impacted by the oil in 1986 and displayed relatively little recovery during the 1987 growing season. The large differences in toxicity between the creek edge and midmarsh zones appeared to be related to the inability of S. alterniflora growing in the physiologically stressful mid marsh zone to tolerate the additional stress of oil toxicity. Oil impacts in the high marsh zone (dominated by S. patens) were intermediate between those of the creek edge and midmarsh zones. Significant negative impacts were noted for most of the study parameters in 1986, however, the heavy mortality observed in the midmarsh zone was not found there. Data from the 1987 growing season revealed relatively little recovery. The creek edge dispersant treatment had severe negative effects on this community in 1986, however, rapid recovery was observed in 1987.

Biomass and species cover data for 1986 and 1987 revealed similar successional trends for the three vegetation zones following the application of oil or dispersant. Annual species, the most abundant of which were Salicornia europaea and Suaeda maritima, increased in abundance in plots where interspecific competition had been reduced by oil or dispersant induced mortality of Spartina. Based on current rates of recovery, the creek edge zone recovered from oiling within one year of treatment and will probably recover from the dispersant treatment within two years. The high marsh zone will probably require three to four years for full recovery, while the midmarsh zone will probably require at least five years.

Résumé

Cette étude décrit les effets d'un épandage expérimental de pétrole brut "Alberta sweet blend" dans la zone du bord de l'anse et dans les zones moyenne et supérieure d'un marais salant de Nouvelle-Écosse, pendant deux saisons de croissance de la végétation. Cette étude décrit également l'impact de l'application du dispersant Corexit 9527 dans la zone du bord de l'anse. Les effets de l'application du pétrole et du dispersant ont été mis en évidence en observant l'évolution de plusieurs paramètres parmi lesquels on peut citer: le taux de croissance des plantes, la densité des tiges, la biomasse, le recouvrement spécifique, la fluorescence des plantes mesurée par induction, les différences anatomiques et les variations du profil du sol. Ces données révèlent des variations notables de la sensibilité des trois zones de végétation au traitement par le pétrole. Ce traitement a peu d'effet dans la zone du bord de l'anse (dominée par Spartina alterniflora à longue tige) durant la saison de croissance 1986 et aucun effet résiduel ne persiste en 1987. La zone du marais moyen (dominée par S. alterniflora a tiges courtes) est au contraire fortement affectée par l'application de pétrole en 1986 et ne montre qu'un faible taux de récupération en 1987. La grande différence qui existe entre la zone du bord de l'anse et celle du marais moyen s'explique par la faible tolérance au stress de S. alterniflora déjà exposée à des conditions physiologiquement stressantes dans le marais moyen. L'impact de l'épandage de pétrole dans la zone supérieure du marais (dominée par S. patens) se situe a un niveau intermédiaire entre celui de la zone du bord de l'anse et celui du marais moyen. Des impacts significativement négatifs ont été mis en évidence pour la plupart des paramètres étudiés en 1986, bien que les forts taux de mortalité observés dans la zone du marais moyen ne se retrouvent pas dans la zone supérieure du marais. Les données de 1987 y révèlent un faible taux de récupération. Le traitement au dispersant produisent des effets négatifs très sérieux dans la zone du bord de l'anse, mais celle-ci récupère rapidement en 1987.

Après l'application de pétrole et de dispersant, les données sur la biomasse et le recouvrement spécifique présentent des tendances similaires en 1986 et en 1987 pour les trois zones de végétation. L'abondance des espèces annuelles, parmi lesquelles les plus abondantes sont Salicornia europaea et Suaeda maritima, augmente dans les endroits ou la compétition entre espèces diminue par suite de la mortalité de Spartina due à l'application de pétrole ou de dispersant. En se basant sur les taux actuels de récupération, la zone du bord de l'anse se remet d'une exposition au pétrole en un an, et d'une exposition au dispersant seul, en deux ans. Une période de récupération de 3 a 4 ans est probablement nécessaire pour la zone supérieure du marais après un épandage de pétrole et une periode de 5 ans est probablement nécessaire pour la zone du marais moyen.

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INTRODUCTION

Salt marshes are highly productive ecosystems which are of great ecological and economic importance. Salt marshes fix large quantitites of carbon, a large proportion of which is exported to estuarine and coastal waters where it fuels marine food webs. Many species of invertebrates, fish and birds utilize salt marshes as feeding, breeding and resting areas. Most of the salt marshes along the Atlantic coast of North America are dominated by the salt marsh cord grass (Spartina alterniflora Loisel). This species is tolerant of high salinity and anoxic conditions in the sediment, yet is capable of very high rates of primary productivity.

Salt marshes are highly susceptable to oil spills since they are regularly inundated by seawater and have low self cleaning potential as a result of the low energy nature of the areas in which they develop. Much variation has been found in the sensitivity of salt marshes to oil spills. Oil toxicity depends on a number of factors including type of oil, the degree of oil weathering, amount spilled, frequency of multiple spill events, time of the year in which the spill occurred, the types of organisms exposed to the oil and the amount of stress experienced by these organisms prior to the oil spill. These factors make it difficult to implement proper mitigative measures when salt marshes are threatened by oil spills. This situation is further complicated by the fact that most salt marsh clean up techniques are ineffective or cause as much if not more damage than the oil (Vandermeulen and Jotcham, 1986). No intervention may be a valid option provided the impact of the oil spill is less than that of the potential clean up option. Evaluation of this option requires the development of oil spill impact studies in a variety of salt marsh communities using different types and amounts of oil applied at different times of the year and monitored over a number of years. This is beyond the scope of most investigations, however, useful information can be derived from a variety of smaller studies.

The dispersal of incoming slicks offers a means of reducing the amount of oil entering the marsh. Generally the use of dispersants in shallow near shore waters is not recommended but may be attempted if it will reduce the amount of oil stranding in the marsh (API, 1985). Near shore applications of dispersant increase the likelihood of dispersant contamination of salt marsh vegetation. Few studies have investigated the impacts of dispersants on salt marsh plant communities (Baker et al, 1984; Delaune et al., 1984 and Smith et al, 1984). These studies have demonstrated that the phytotoxicity of various dispersant formulations varies considerably. It is therefore important to document the toxicity of a variety of dispersants

in order to determine which may be safely used in close proximity to salt marsh vegetation.

In this study, the impacts of Alberta sweet blend crude oil and the dispersant Corexit 9527 were studied in a Nova Scotia salt marsh over the course of two growing seasons. The impacts of experimental crude oil spills were investigated in creek edge, midmarsh and high marsh vegetation zones while dispersant impacts were studied only in the creek edge zone. Oil and dispersant impacts were studied at the community, species and cellular levels of biological organization in order to provide a comprehensive approach to evaluation of the biological effects of the treatments.

This is a continuation of a study initiated in 1986 (Lane et al., 1987) and presents data collected during both the 1986 and 1987 growing seasons following the oil and dispersant applications in 1986. More detailed descriptions of treatment impacts on vascular plant parameters during the 1986 growing season and effects of oil and dispersants on salt marsh algal and bacterial communities are available in that report. In this study, attention was focused on parameters from the 1986 study in which recovery was not evident at the end of the 1986 growing season.

MATERIALS AND METHODS

DESCRIPTION OF STUDY SITE

The field study site was located at Conrods Beach on Petpeswick Inlet ($44^{o}42'N$, $63^{o}11'W$) (Figure 1). The Conrods Beach salt marsh has developed behind a barrier dune system, and is drained by a single channel which penetrates the dune system. The vegetation of the salt marsh is divisible into three distinct zones, which will be referred to as creek edge, midmarsh, and high marsh zones.

The creek edge zone was dominated by a lush growth of Spartina alterniflora. Small quantities of Salicornia europaea, Plantago juncoides, Suaeda maritima, and Atriplex patula were also present in this zone. The creek edge zone was usually restricted to within two metres of drainage channels in the marsh. The lush growth of Spartina alterniflora was attributable to the relatively well drained sediments of this zone.

The midmarsh zone was dominated by short Spartina alterniflora, with lower abundances of Spartina patens, Salicornia europaea, Suaeda maritima, Triglochin elata, Limonium nashii, and Plantago juncoides. This was the most extensive of the three zones in the Conrods Beach salt marsh and was generally found in poorly drained areas in the marsh interior. The stunted growth of Spartina alterniflora in the midmarsh zone is generally associated with extremely low redox potential of the sediments and high salinity of the interstitial water of the sediments of this zone.

Spartina patens was the dominant species of the high marsh zone. Small quantities of Spartina alterniflora, Salicornia europaea, Triglochin elata, Suaeda maritima, and Glaux maritima were also found in association with S. patens. The high marsh zone was located on slightly elevated, better drained and aerated soils of the marsh.

EXPERIMENTAL DESIGN

A total of twenty-one 0.4m x 4.0 m ($2.m^{2}$) plots were established at random locations in the salt marsh. Nine plots were positioned in the creek edge zone, with three replicate plots assigned to each of three treatments (control, oil and dispersant). In the midmarsh and high marsh zones, only control and oil treatments were assigned for a total of six plots per zone. Each plot was separated from other plots by a 10 m buffer zone to prevent or reduce cross contamination by either oil or dispersant from other plots. Nondestructive sampling was conducted within the interior of the plots

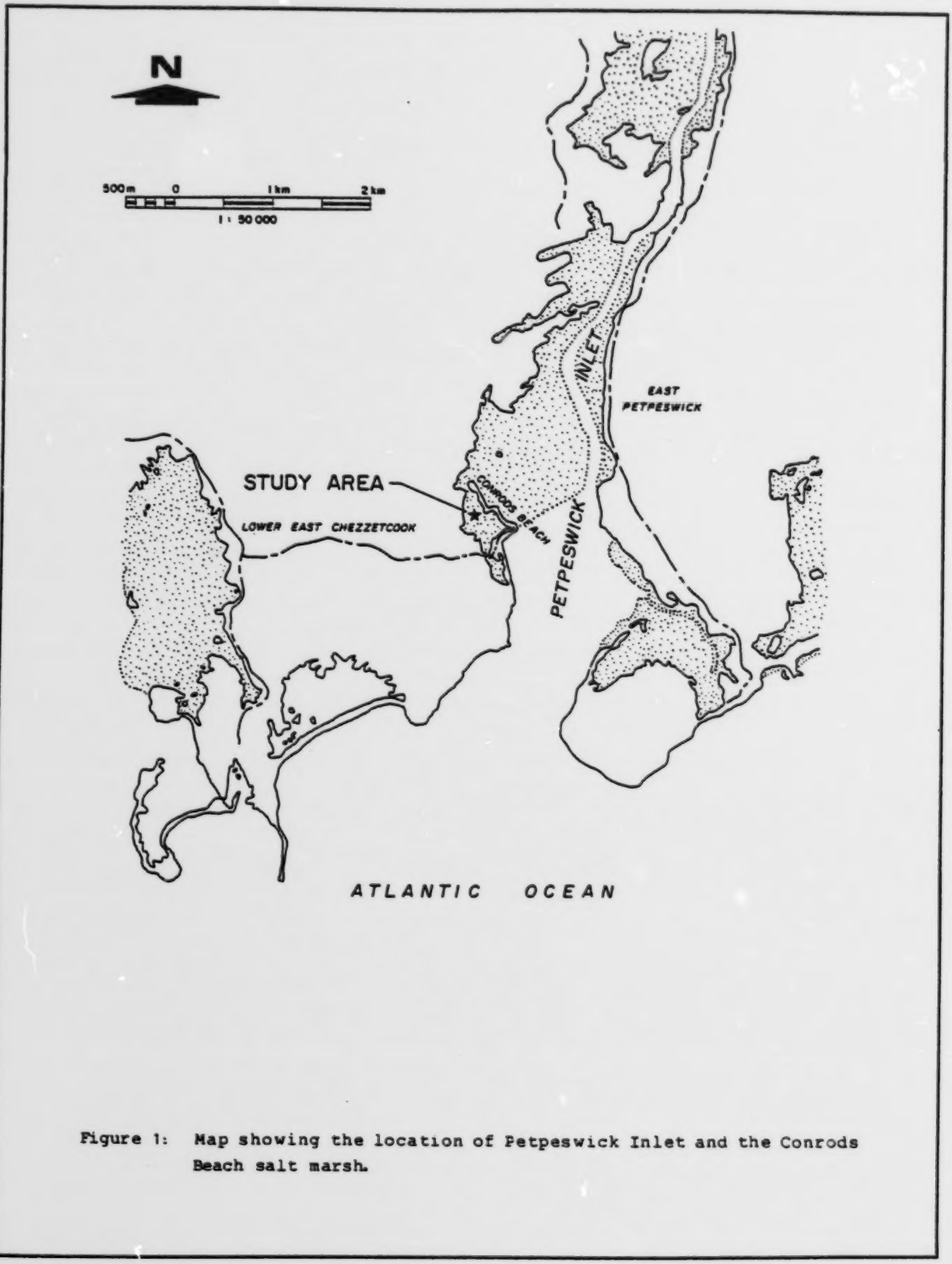


Figure 1: Map showing the location of Petpeswick Inlet and the Conrods Beach salt marsh.

while destructive sampling was done in a 0.25m wide strip surrounding each plot which received the same treatment as the plot interior. Each quadrat was surrounded by a wall of oil sorbent material to prevent the escape of oil from the quadrat. Quadrats were also covered with garden netting to exclude wildlife. Field measurements commenced on May 9 (day 1) and treatments were applied on July 7 (day 60) in 1986.

In the 1986 study three treatments were applied to each of the three vegetation zones. These included oil alone, dispersant alone and oil and dispersent applied to the same plot. Funding constraints in 1987 resulted in a reduction in the number of plots monitored during the 1987 field reason. Only the control and oiled plots were monitored in all vegetation zones along with dispersant treated plots in the creek edge zone.

OIL AND DISPERSANT APPLICATIONS

Alberta sweet blend crude oil, weathered by evaporation and stirring for 24 hours to 15-25% loss by weight, was used for all experiments. In the field experiments, 2.25 liters of weathered oil per plot were applied from a back-pack sprayer approximately two hours after the onset of the ebb period yielding a nominal 0.5 mm oil slick. The tide did not rise high enough to cover the marsh surface with water on the day of treatment applications, therefore, each plot was sprayed with seawater prior to treatment. The oil spray application was done so that the spray was held near the marsh sediment surface, at about half of the mean plant height, thereby simulating contamination by a surface oil slick.

For the dispersant treatment, ethylene glycol-based Corexit 9527 was mixed with water in a ratio of 1 part dispersant to 10 parts seawater. A total of 2.25 liters of the dispersant:seawater mixture was applied to each plot from a back-pack sprayer. Application differed from that of the oil application, in that the dispersant spray was directed at the vegetation from 50 cm above the sediment surface. This application was intended to mimic a near shore application of dispersant on an incoming tide.

PHYSICAL AND CHEMICAL PARAMETERS

Soil Profile Descriptions

Soil profile descriptions were recorded for all cores taken for chemical analysis. On each sampling date, in each vegetation zone, one core was taken from each of the 3 replicate oil plots. During the 1986 field season only one core was taken from each of the control plots, however, during the 1987 field season cores were taken from all control replicates. At each

sampling site, a core 6 cm wide by 15 cm long was removed with a modified bulb planter. Each core was split longitudinally and the thicknesses of distinctive sediment layers and presence of oxidizing and reducing zones were recorded on a standard core diagram. The presence of bacterial zones, algal mat communities, the colour of roots, and the degree of root mat development were also recorded for each core. In 1986, cores were taken on five dates between early July and early September, while in 1987, cores were taken on three dates between late June and late August.

Soil Chemistry

Using the cores collected for soil profile descriptions, a 50 g sample was removed from each core and was extracted 3 times with 20 ml of methylene chloride. The resultant solution was dried, taken up in hexane and cleaned up. Samples were cleaned by pouring them through a mini-column containing sodium sulfate, copper and activated florisil. Total oil and grease values were determined with UV fluorescence analysis, scanning from 200 to 300 nm. The data were expressed as ppm of chrysene equivalents (UV fluorescence at 256 nm).

Selected sample extracts were analysed by glass capillary gas chromatography using a flame ionization detector. Relative concentrations of the n-alkanes were determined from the chromatograms. Identifications of the peaks were made using glass capillary gas chromatography mass spectrometry. Procedures followed those of Geiger and Schaffner (1978).

VASCULAR PLANT PARAMETERS

Plant Height

Within each plot, 40 Spartina shoots were systematically selected at 10 cm intervals and their heights were measured. Plant height was measured as the distance from the tallest point of the plant to ground level. Measurements were taken on eight occasions during the growing season in 1986 and on five occasions during the 1987 growing season.

Stem Density

Within each plot, 12 miniquadrats were systematically positioned at 30 cm intervals. In Spartina alterniflora dominated marsh zones (creek edge and midmarsh zones), $0.01 \cdot m^{2}$ quadrats were used while in the S. patens dominated zone (high marsh zone), $0.006 \cdot m^{2}$ quadrats were used. All shoots in each quadrat and all flowering shoots in each plot were counted and means and standard deviations were calculated for each treatment in each

vegetation zone on each sampling date. Measurements were taken on four occasions during the 1986 growing season and on five occasions during the 1987 growing season.

Biomass

Biomass harvesting was conducted in the first week of September in both 1986 and 1987. A 0.09$^{\circ}m^{2}$ quadrat was systematically positioned in one end of each plot and all aboveground living biomass in the quadrat was harvested, sorted by species, dried at 80° C in a convection oven and weighed on an analytical balance. Standing dead biomass was harvested, dried and weighed but was not sorted by species. All flowering stems in each plot were harvested. Reproductive portions were removed from vegetative portions and weighed separately. The mean Spartina shoot weight was calculated by dividing the biomass of Spartina alterniflora (creek edge and mid marsh) or Spartina patens (high marsh) in the plot by the mean stem density of the plot recorded on the day that the biomass was harvested. Mean above-ground standing crops for living plants, standing dead, reproductive tissues and mean shoot weight were calculated for each treatment in each vegetation zone for each year.

Species Cover

Percent cover for each species of vascular plant was estimated in each plot. Cover values were estimated relative to soil surface area rather than to other species in the quadrat; therefore, total cover could be higher than 100% when species overlapped. Mean cover values were calculated for each species in each treatment in all vegetation zones for three sampling dates in 1986 and five sampling dates in 1987.

Fluorometry

An explanation of the principle of plant fluorescence induction measurements is presented in Appendix 1. Three plants were randomly selected from each plot for fluorometric analysis. The upper-most fully expanded leaf was removed. All of the leaves from a particular plot were placed in a plastic bag into which distilled water was sprayed. The samples were then placed in a light proof box for transport to the laboratory. Harvesting and preparation of the leaves required approximately 0.5 hours. During transport the leaves were allowed to equilibrate in the dark for 1.5 hours. Working illumination in the laboratory was provided by a 6 volt lantern fitted with a green filter. Fluorometric measurements were made with a Brancker Research Ltd. (Ottawa) model SF-20 portable fluorometer. The fluorescence induction curve was

monitored for 100 seconds. Initial, peak, transient peak, and final readings were recorded (Figure 2). Values used in analysis included the peak variable fluorescence (peak reading minus initial reading) and the 100 second difference value (value at 100 seconds minus initial value).

Leaf Anatomy

In mid-July in 1986 and 1987, two leaves were randomly selected from each plot. These leaves were pooled by treatment within each zone and three were randomly chosen from each treatment for observation. Radial cross-sections were taken from areas on each leaf exhibiting different symptoms of the treatments, and were examined under a microscope at 200x. Observations included the abundance, relative sizes, and coloration of chloroplasts, measurements of cuticle thickness and the presence or absence of plasmolysis.

STATISTICAL ANALYSIS

Statistical analyses were conducted on plant height, stem density, above ground biomass for several species, reproductive and mean shoot biomass, cover, fluorometry parameters, reducing zone thicknesses and total oil and grease concentrations. In most cases, both 1986 and 1987 data were analysed. Transformations as determined by tests for normality and homoscedasticity in 1986 were continued. Data analysis was conducted using the SPSS/PC+ V2.0 statistical package (Norusis, 1988a, 1988b). Graphs were produced with SPSS/U.C.+ Graphics (SPSS Inc., 1986).

Preliminary analysis consisted of both general descriptive statistics and analysis of variance to test whether replication was a factor. Subsequent analysis investigated the effects of treatments and time (year, and where appropriate, visit). The principle investigative technique was analysis of variance. The number of levels and the interactions tested varied with the particular data set.

Height data was transformed as follows:

$$y = \ln ((x/14) + 1).$$

where 14 was the overall median height for the 1986 data.

Density was also ln transformed:

$$y = \ln (x + 1).$$

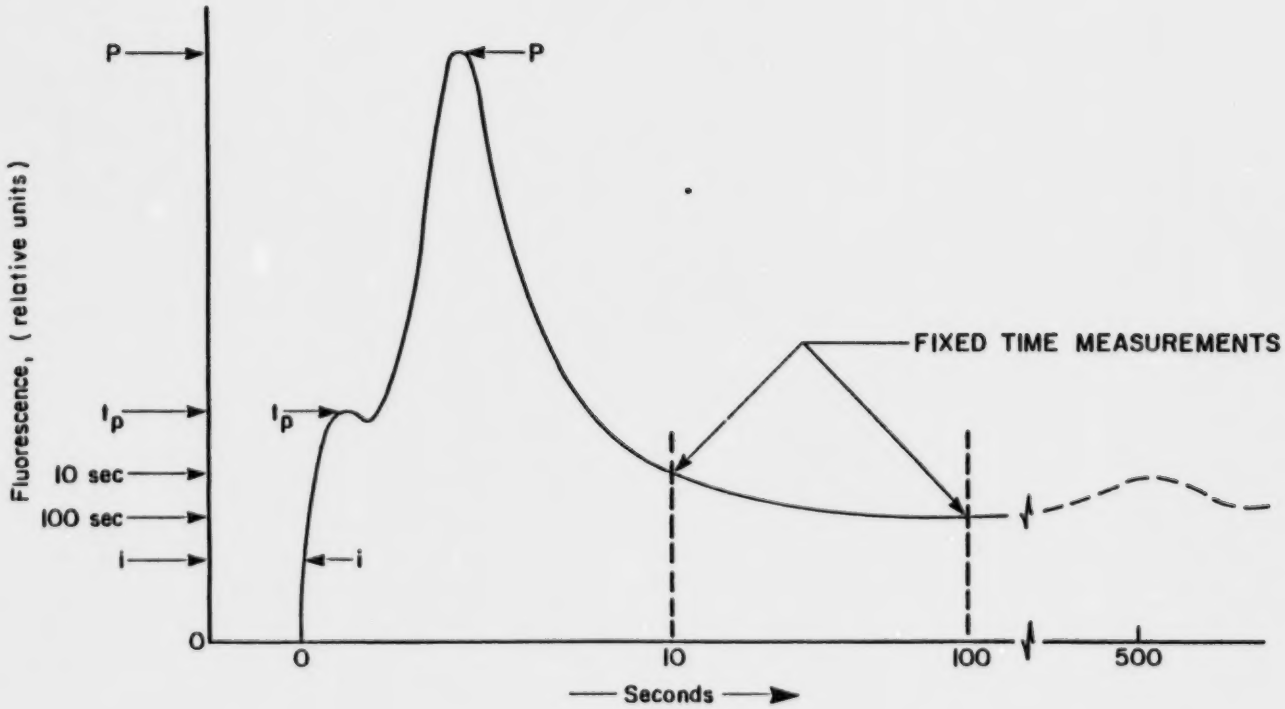


Figure 2. Diagramatic representation of the fluorescence induction curve for Spartina alterniflora. Arrows indicate recorded fluorescence values. The initial value (i) is the "fixed fluorescence". Fluorescence above this level is "variable fluorescence" which is sensitive to the physiological status of the photosynthetic system. We refer to (p-i) as "peak variable fluorescence" and 100 second values minus initial value as the "100 second differences values". tp refers to a transient peak.

Square root transformations were used on both cover and biomass data. Statistical analysis of the biomass data reported here was restricted to the two Spartina spp., Salicornia europaea, Suaeda maritima, total above ground living biomass and standing dead biomass. Several subsets of the dominant Spartina species were also investigated, including flower weight and weight per shoot.

Cover data for the creek edge, midmarsh, and high marsh were entered into discriminant analyses. The results of 4 estimates for each plot on each day were incorporated. The estimates were divided into groups according to treatment. All cover estimates from the first sampling date in 1986 were coded as controls since they preceeded treatment applications. Separate ordinations were conducted including and excluding standing dead biomass.

RESULTS AND DISCUSSION

PHYSICAL AND CHEMICAL EFFECTS

Soil Profile Descriptions

Sulfate reduction activity is the major determinant of soil profile patterns in salt marshes. Water logging of the sediments combined with microbial respiration create anoxic conditions under which sulfate reducing bacteria may proliferate. These bacteria utilize sulfate as an electron acceptor in the anaerobic respiration of small molecular weight organic compounds. It is generally believed that the carbon sources utilized by sulfate reducing bacteria are waste products of the fermentation of detritus. The reduction of sulfate produces hydrogen sulfide which is stable under anaerobic conditions. Hydrogen sulfide may also react with iron to form ferrous sulfide and eventually pyrite (Weibe et al., 1981). Deposition of the black iron sulfides produces a grey or black colour in the sediments. In zones where the sediments are oxidized, the sediment color is brown as a result of the oxidized state of the iron compounds. The transition zone between brown and grey sediments is characterized by a decline in redox potential from positive to negative values (Jorgensen, 1983). Sediment colour may, therefore, serve as an indirect indicator of sulfate reduction activity which in turn indicates the presence of anaerobic conditions.

Soil profile descriptions were performed during both the 1986 and 1987 field reasons, however, during 1986 only a single control plot was sampled on each sampling date. Consequently, only data from 1987 was suitable for statistical analysis. No data were collected for dispersant treated plots. Figure 3 presents the mean thickness of reducing zones in control and oiled plots in each of the vegetation zones in 1987. All three zones exhibited similar trends over the course of the growing season. Reducing zone thickness in control plots declined over the course of the growing season while that of oil treated plots increased. Reducing zone thickness in the oiled plots was initially lower than controls at the beginning of the growing season (Day 45) but increased to control levels by the end of the growing season (Day 110). In the creek edge plots on day 45, reducing zones were only 61% as thick as in control plots, however, this difference was not statistically significant. By day 75, reducing zones in the oiled plots had thicknesses similar to those of the control plots.

In the midmarsh zone mean reducing zone thickness in oiled plots was 53% of control plot values on day 45 and 51% of control values on day 75. These

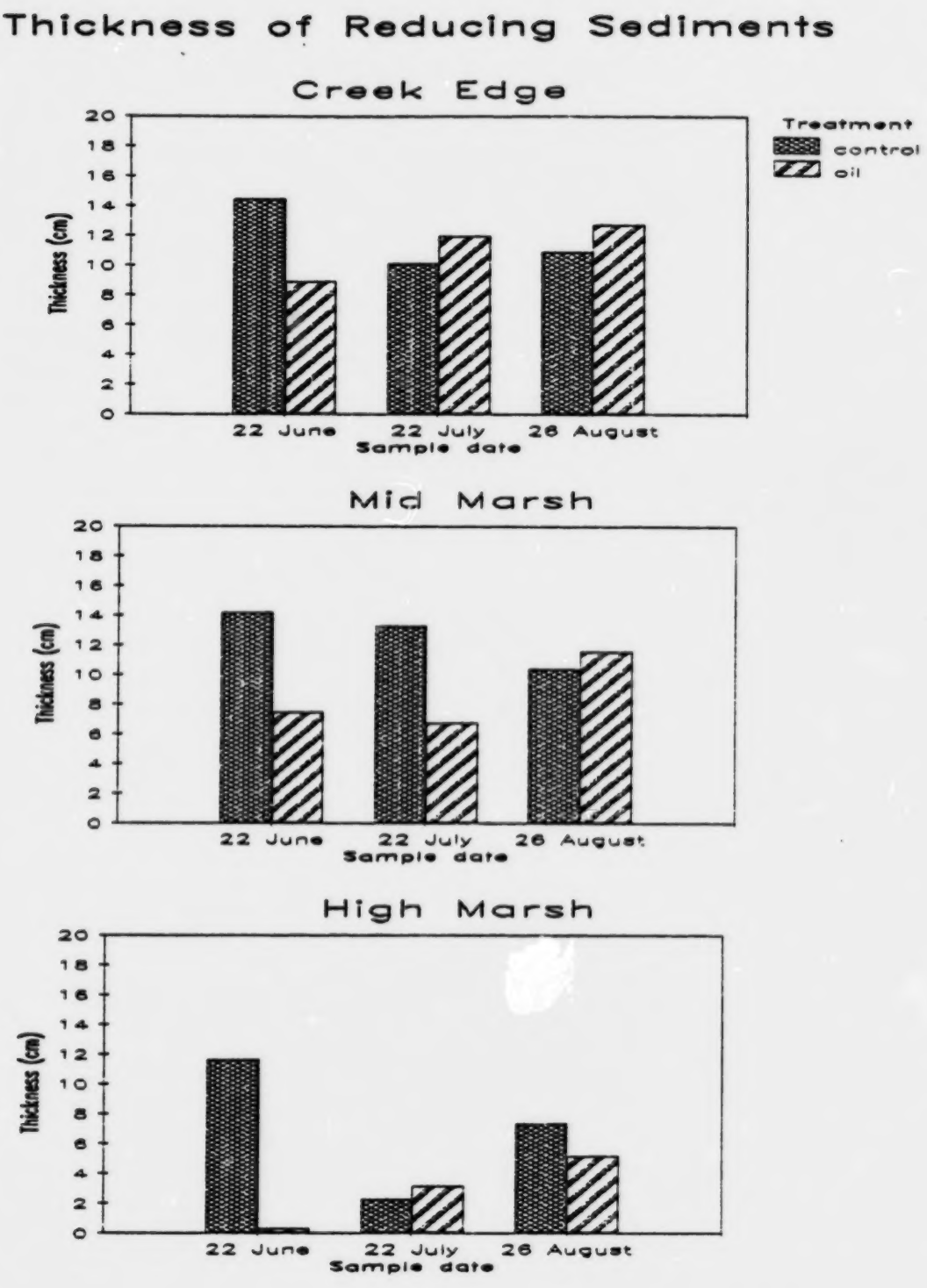


Figure 3: Histograms of mean reducing zone thickness in the creek edge, midmarsh and high marsh zones in 1987. Oil was applied on day 60.

differences, however, were not significant on either sampling date. Reducing zone thickness for oiled and control plots were similar on day 110.

Reducing zone thickness in oiled high marsh plots was significantly lower than control plots on day 45 (p 0.001, ANOVA) but returned to control values by day 75.

Contamination of salt marsh sediments by crude oil could affect reducing zone thickness in several ways:

1) An oil layer on the sediment surface or high concentrations in the sediment could provide a barrier to diffusion of oxygen into the sediments resulting in an increase in reducing zone thickness in oiled plots.

2) Oil induced mortality of Spartina plants could reduce the degree of sediment oxidation. Howes et al (1981) have demonstrated that growth of Spartina alterniflora was associated with increases in sediment redox potential. Spartina plants supply oxygen to the rhizosphere by means of passive diffusion of air through continuous gas spaces from shoots to rhizomes and roots and possibly by means of the exudation metabolically produced oxidants such as hydrogen peroxide into the rhizosphere. Mortality of the Spartina sward might be expected to be associated with an increase in reducing zone thickness in the oiled plots.

3) The crude oil could serve as a substrate for oil degrading bacteria. Respiration by these bacteria could decrease the depth to which oxygen can penetrate the sediments, resulting in an increase in reducing zone thickness in the oiled plots.

4) The crude oil could poison populations of heterotrophic bacteria in the sediment causing a reduction in bacterial respiration, increasing the depth to which oxygen could penetrate the sediments. This could cause a decrease in the thickness of reducing zones in the oiled plots.

The evidence presented in figure 3 suggests that the oil treatments were responsible for a long term (approximately one year) increase in soil oxidation, possibly as a result of a reduction in microbial respiration rates caused by hydrocarbon toxicity. Unfortunately, no estimates of microbial respiration were available for the field experiment, so there is no direct evidence to support this hypothesis. There are, however, several

pieces of circumstantial evidence which support this hypothesis. Laboratory experiments in 1986 (Lane et al, 1987) using midmarsh sediment from the study area showed that Alberta Sweet Blend crude oil significantly reduced sediment respiration, although only for a period of ten days. The effects may have been more persistent under the harsher conditions experienced in the field (cooler temperatures and constant waterlogging. Standing dead biomass data for 1986 (see page 19) indicated that decomposition rates had been substantially reduced by the oil treatment. Rates of decomposition appeared to have returned to normal by the end of the 1987 growing season.

Other aspects of the sediment profiles did not change as a result of oil applications. Root development differed between vegetation zones but was similar between treatments within zones. The high marsh zone (dominated by Spartina patens) and midmarsh zone (dominated by S. alterniflora) had well developed mats of fine roots which were largely concentrated near the sediment surface, presumably to avoid high concentrations of toxic hydrogen sulfide and other physiological constraints associated with the anaerobic sediments in these zones. In the creek edge zone (dominated by S. alterniflora) there were fewer fine roots and more large rhizomes which extended into the sediments somewhat deeper than in the previous two vegetation zones. Frequent exchange of interstitial water in this zone would reduce the concentrations of toxic hydrogen sulfide, allowing deeper penetration of root systems.

Sediments in contact with the fine root mats of the midmarsh and high marsh plots were generally oxidized, however, oxidized zones were absent in the creek edge quadrats where fine roots were relatively scarce. No apparent declines in the extent of these zones were associated with the oil treatments in either 1986 or 1987.

Soil Chemistry

Figures 4 - 6 present hydrocarbon concentrations in oil treated and control plots in the three vegetation zones in 1986 and 1987. There was considerable variation between replicates on the various sampling dates. This could be caused by several factors. Initial distribution of the oil within the plots may have been uneven resulting in zones of higher or lower hydrocarbon concentrations. Lipophobic conditions in the water saturated sediment may have led to the concentration of oil in low areas within the plots. Oil draining down grass shoots may have penetrated into the sediment as a film around plant roots and rhizomes causing localized concentrations of oil in the sediment.

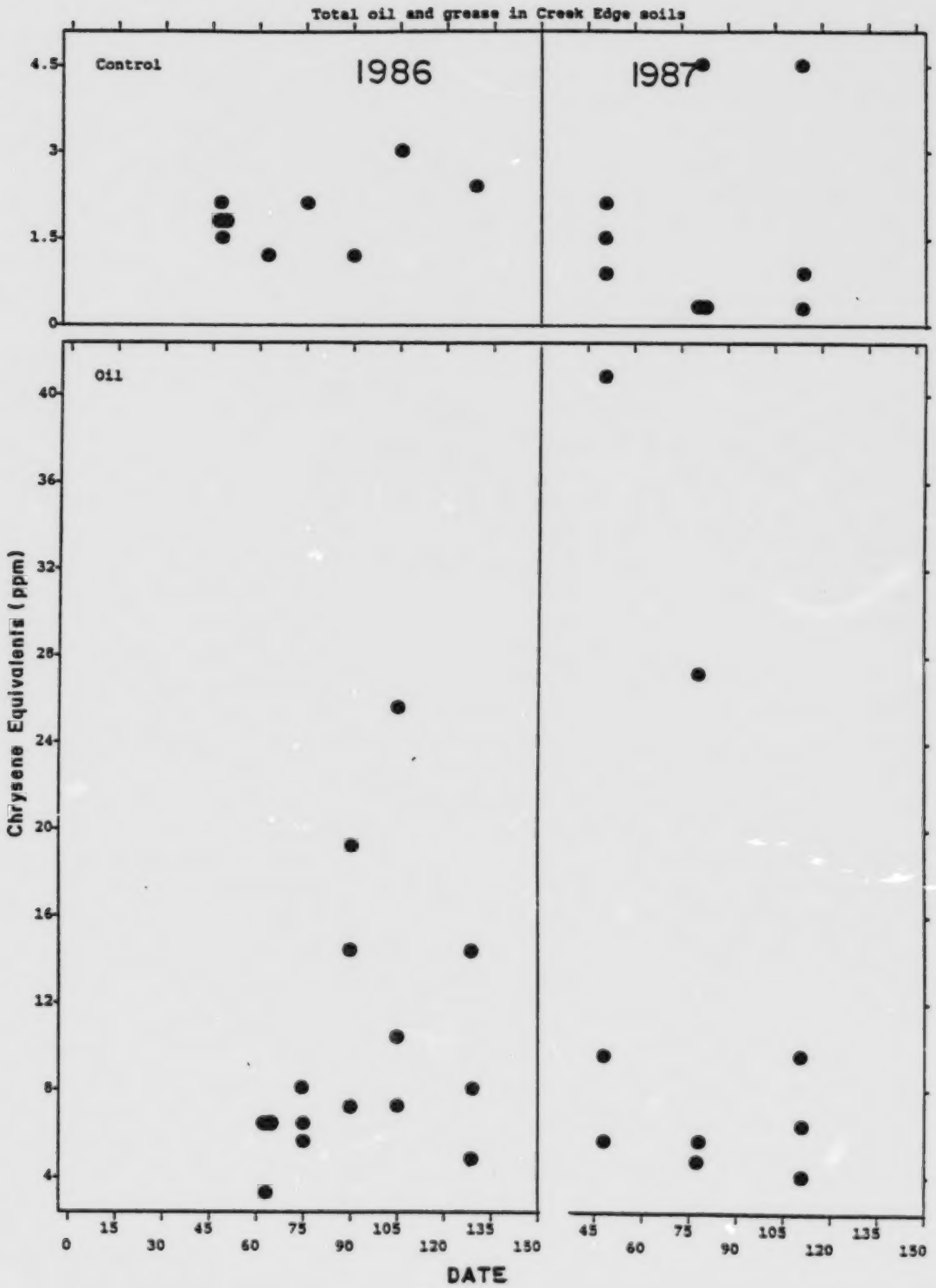


Figure 4: Scatter plot of total oil and grease concentrations in control and oil treated plots in the creek edge zone in 1986 and 1987. Oil was applied on day 60 , 1986.

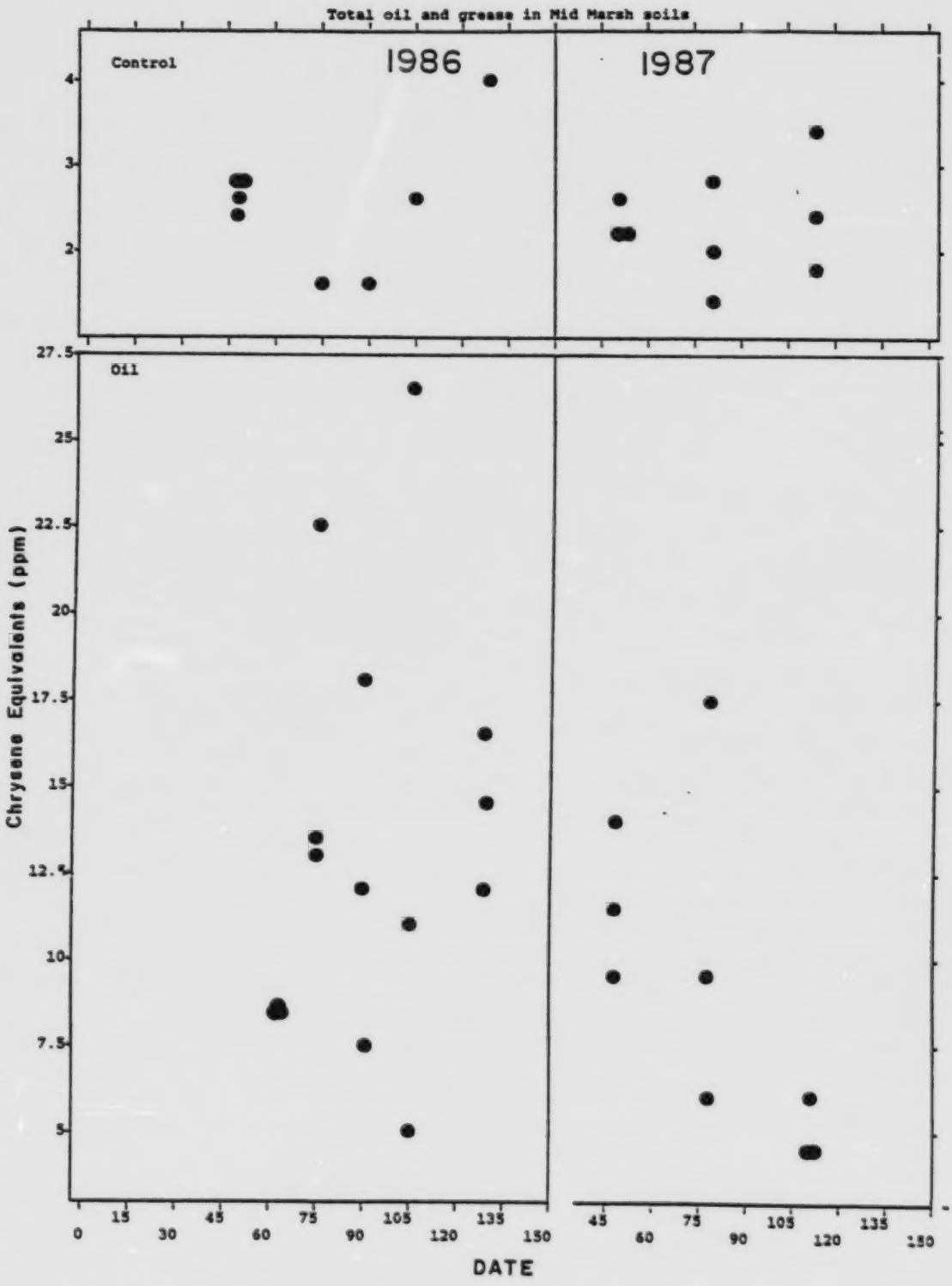


Figure 5: Scatter plots of total oil and grease concentrations in control and oil treated plots in the midmarsh zone in 1986 and 1987. Oil was applied on day 60., 1986.

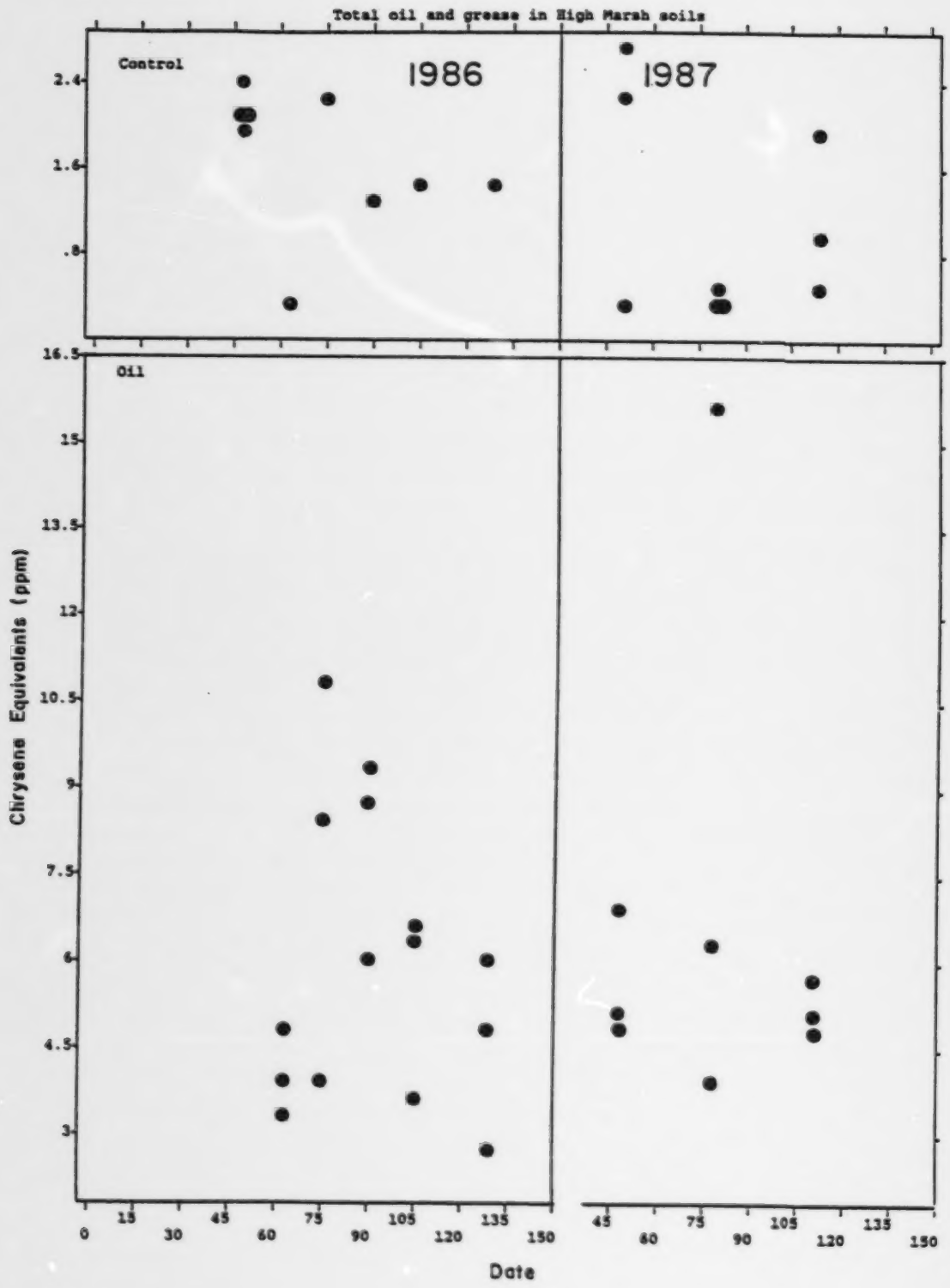


Figure 6: Scatter plots of total oil and grease concentrations in control and oil treated plots in the high marsh zone in 1986 and 1987. Oil was applied on day 60 , 1986.

The data are too variable to draw definite conclusions about the fate of the oil in the various vegetation zones other than to demonstrate that there were substantially more hydrocarbons in the oiled plots than in the control plots during both the 1986 and 1987 growing seasons. There were, however, some general trends in the data. In all zones hydrocarbon concentrations were initially low two days after oil applications but reached maximum concentrations within 30 days of application after which concentrations appeared to gradually decline. The initial increase in hydrocarbon concentrations was probably attributable to several factors:

1) Oil deposited on leaves may have flowed down the stems and onto the sediment over the course of several days.

2) Tidal inundation may have lifted oil off of plants and deposited it on the sediment surface.

3) Oil killed plants or oiled detritus laying on the surface of the sediment may have been incorporated into the sediment.

Hydrocarbon concentrations in the sediment were generally high in the creek edge and midmarsh zones and low in the high marsh zone. Low oil concentrations in the high marsh zone were almost certainly attributable to interception of the sprayed oil by the large standing crop of dead biomass in this zone. In many plots standing dead biomass was up to twice as high as the living biomass, suggesting that Spartina patens was highly resistant to decay. During the 1987 field season oil stained dead stems were still common in the oiled plots. Oil absorbed by the standing dead biomass would not be available for incorporation into the sediment for many months. In addition, this oil would be expected to weather much faster than oil incorporated into the sediments. Maintenance of a high standing crop of dead biomass may have been a factor limiting the impact of oil in the high marsh zone.

Gas chromatography of oiled marsh sediment samples revealed that the rate of weathering of the oil in the sediments was highly variable. By the end of the 1986 field season oil in the sediments on average had weathered to the point where all hydrocarbons under C-10 had been lost. This corresponded to a volume loss of 33% in an oil evaporation experiment conducted in the laboratory. The evaporation experiment determined that evaporative loss of the oil stabilized at 36% after two months of weathering (Table 1), suggesting that evaporative losses were nearly complete in the field. Further losses would be attributable to flushing of oil from the sediments and microbial degradation. By the end of the 1987 growing season hydrocarbons below C-14 had been weathered from the oil and

Table 1. Oil evaporation experiment: Percent of original oil remaining at specified times. Ten grams of oil were weathered in an open container (6 cm wide by 1 cm deep) at 20°C.

TIME (HOURS)	PERCENT OF ORIGINAL OIL WEIGHT
0	100
18	81
72	75
187	72
505	67
1,053	65
1,344	64

in two of nine gas chromatography analyses no oil was detectable.

Penetration of oil into the sediments was found to be generally restricted to the upper 15 cm (Table 2). The highest concentrations were found in the 0-5 cm range. Oil penetration was lowest in the midmarsh and high marsh zones where 77 and 76% of the oil in the upper 15 cm of the sediment was found in the upper 5 cm of the sediments. Penetration was somewhat deeper in the creek edge zone with only 61% of the oil contained in the upper 5 cm of the sediments. The deeper oil penetration in the creek edge zone was probably caused by the lower water table in this zone which would permit oil to percolate deeper into the sediments. The lack of a dense root mat in this zone may also have permitted deeper penetration of oil into the creek edge sediments. Hydrocarbon concentrations in the 5-10 cm depth range tended to be slightly elevated above control surface concentrations in the midmarsh and high marsh zones and were up to four times higher than control values in the creek edge zone. Concentrations in the 10-15 cm depth range were comparable to control values at 0-5 cm in all vegetation zones. This corresponds well to the distribution of hydrocarbons in marsh sediment noted by Hershner and Lake (1980). They found no oil below 20 cm five months after multiple dosings of No. 2 fuel oil. Over 90% of the oil was restricted to the upper 10 cm of the sediment. Other studies have found much deeper penetration. Lytle (1975) found oil to a depth of 42 cm in an experimental oil spill and Burns and Teal (1971) found oil to a depth of 70 cm after the West Falmouth oil spill.

Table 2. Mean Sediment Hydrocarbon concentrations at 5 cm depth intervals. Values for 10-15 cm are based on one sample. No data were available for high marsh hydrocarbon concentrations at 10-15 cm.

Total Oil and Grease
(ppm Chrysene equivalents (1 standard deviation))

Treatment	Depth (cm)	Creek Edge	Mid Marsh	High Marsh
Oil	0-5	48.0 (34.9)	24.7 (11.2)	27.6 (4.2)
	5-10	22.2 (28.4)	6.7 (3.9)	8.6 (7.8)
	10-15	8.6	0.6	-
Control	0-5	5.6 (8.8)	5.6 (2.8)	1.8 (2.6)

IMPACTS ON VASCULAR PLANTS

Plant Height

In the creek edge zone in 1986, significant reductions in plant height were noted on all post treatment days for dispersant treated plots (Figure 7 ($p<0.001$, ANOVA)). Plant heights in dispersant treated plots increased substantially between days 102 and 123. This trend was probably caused by selective mortality of short plants which would increase mean plant height, although growth rates of surviving plants probably did not increase. Short plants would be at a disadvantage in competition for light and would, therefore, suffer more stress than the dominant plants. Higher mortality rates would, therefore, be expected for smaller plants. The oil treatment had relatively little impact on plant height in 1986, although oiled plants were significantly shorter than controls on days 89 ($p=0.043$, ANOVA) and 123 ($p=0.034$, ANOVA).

In 1987, there was little or no evidence of oil or dispersant impacts on height growth. Early in the growing season (days 26 and 48), oil and dispersant treated plants were slightly, although significantly, taller than control plants ($p<0.001$, $p=0.021$, ANOVA). This may be attributable to reductions in stem density associated with these treatments which would reduce competition and allow more rapid growth early in the growing season. No significant reductions in plant height were noted for any of the treatments later in the growing season (Appendix 3), with the exception of oiled plants on the last sampling date (day 115) ($p=0.034$, ANOVA).

These data suggest that there was a rapid recovery of plant height growth associated with the dispersant treatment. This is not unexpected since the water miscible dispersant would be rapidly dissipated by tidal action. Data for oiled plots indicated little change in height growth between 1986 and 1987. This suggests that the oil treatment had no short term or long term effects on height growth.

In the midmarsh zone in 1986, oil did not significantly reduce plant height growth (Figure 8, Appendix 3). This was unexpected since heavy mortality was noted in oiled plots. Rapid reduction in stem density of small plants may have maintained mean plant height at an artifically high level during this period. This is supported by the fact that plant heights in oiled plots were significantly shorter than controls throughout 1987 (Appendix 3). It is difficult to determine whether there was any recovery of plant growth between 1986 and 1987 since trends for 1986 were masked by selective mortality of small plants making direct comparsions impossible. Data for

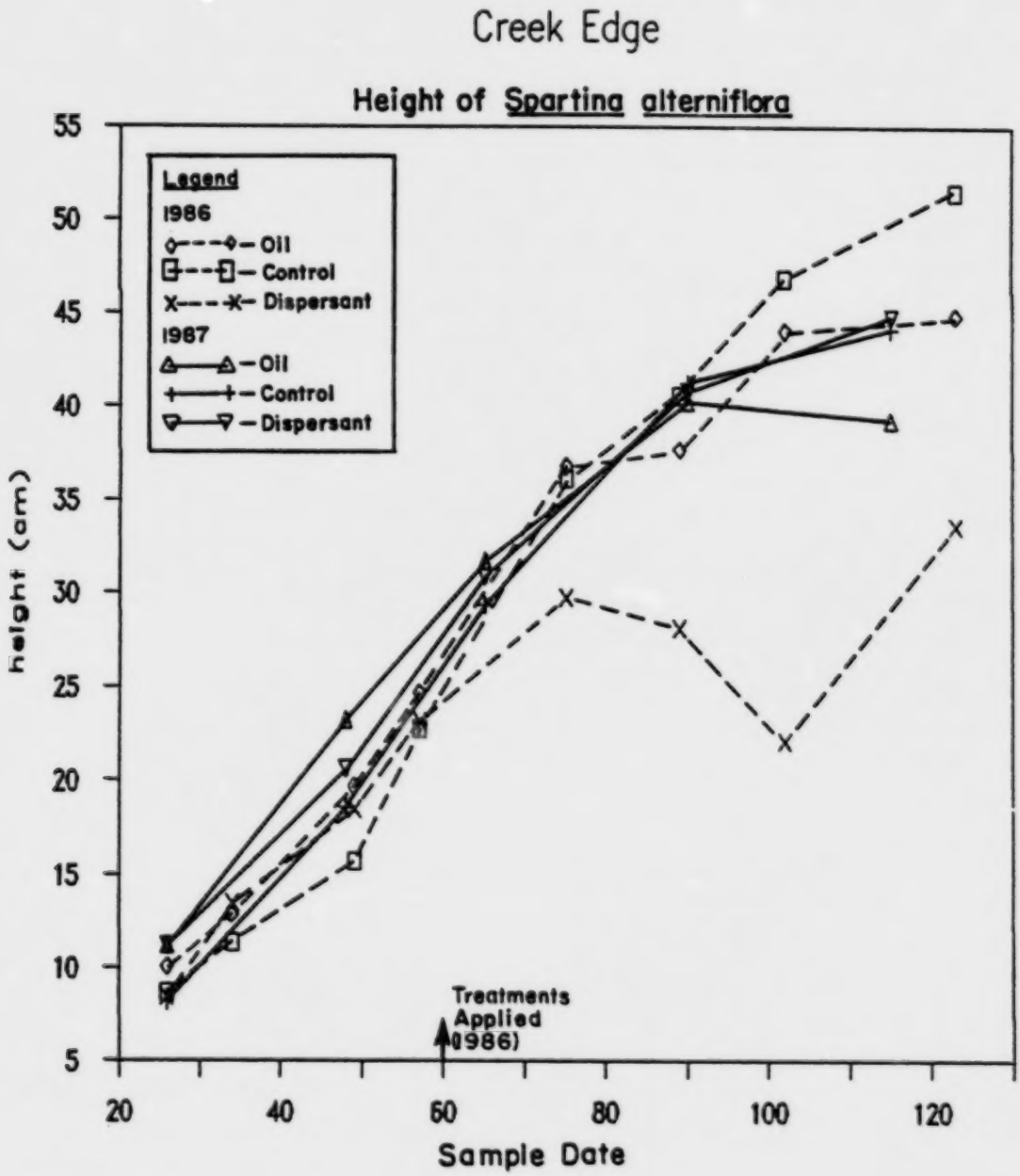


Figure 7. Plot of mean Spartina alterniflora height for the creek edge zone versus time.

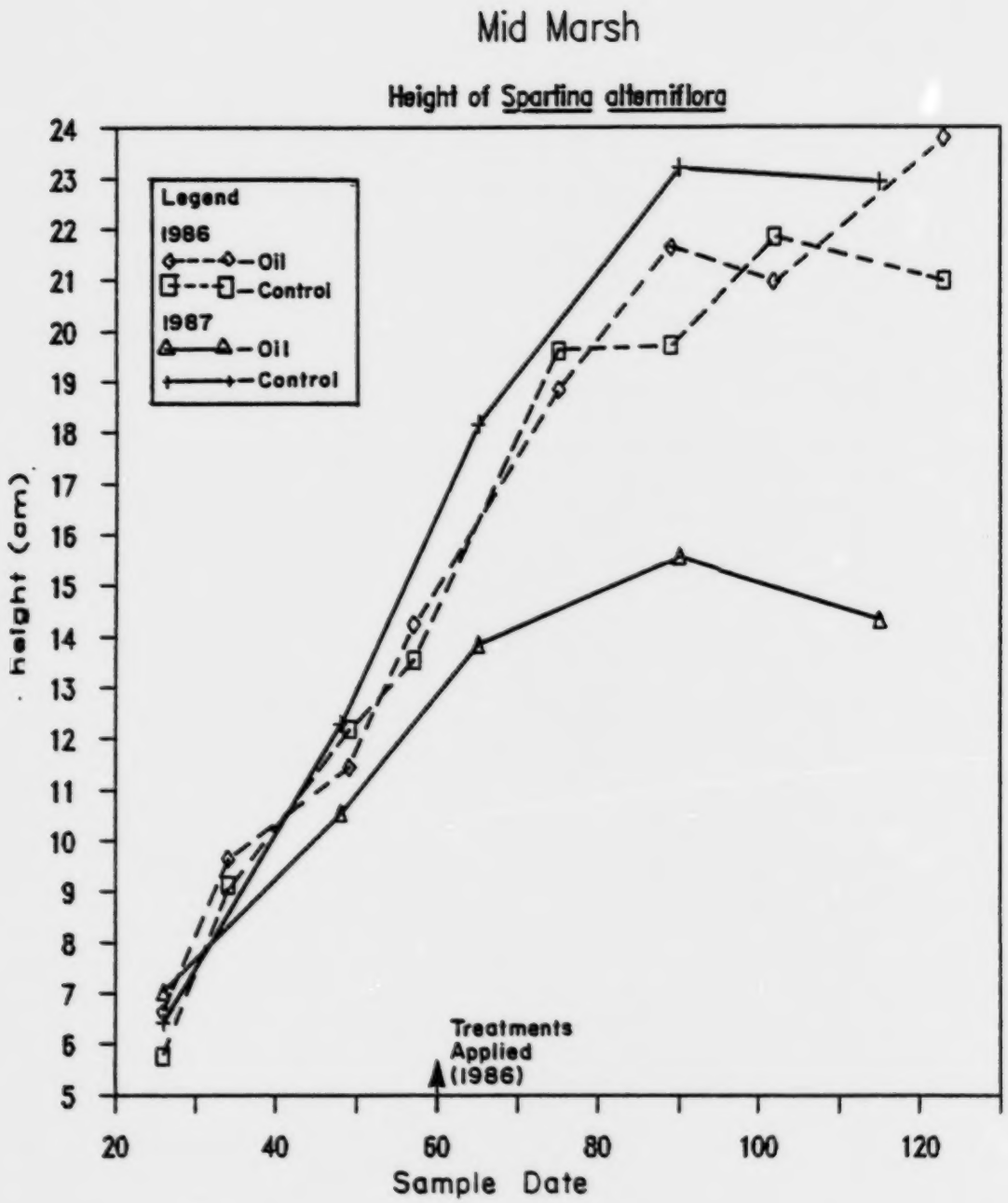


Figure 8: Plot of mean Spartina alterniflora height for the midmarsh zone versus time.

1987, however, indicate that plant height growth was still significantly affected by the oil treatment.

The larger reductions in Spartina alterniflora height in oiled midmarsh plots relative to similarly treated S. alterniflora in the creek edge zone were probably attributable to differences in sediment oxidation status (Eh) between the midmarsh and creek edge zones. Sediment oxidation status is generally lower in the midmarsh zone than in the creek edge zone (Howes et al., 1986) making it a more hostile environment for plants. Low oxygen concentrations lead to decreased ability to take up nitrogen from the sediment (Morris and Dacey, 1984), increased root respiration rates (Mendelssohn et al., 1981) and increased production of hydrogen sulfide which is toxic to root metabolism (Allam and Hollis, 1972). Untreated S. alterniflora growing in the midmarsh zone were shorter than creek edge plants, indicating the stressful nature of the midmarsh zone. The additional stress of oil toxicity was enough to alter the carbon balance of the midmarsh plants (i.e. increased respiration decreased photosyntnesis) to the point wnere height growth was slowed. In the creek edge zone the oil would alter the carbon balance in the same way, however, activities such as flowering which are relatively unimportant to rhizomatous plants such as Spartina would be sacrificed rather than neight growth. In the midmarsh zone, existing stressful conditions would nave already reduced the proportion of photosynthates available for flowering so that additional stress would deplete carbon stores normally devoted to height growtn.

In the high marsh zone in 1986 (Figure 9), height growth of Spartina patens in oiled plots was significantly lower than the heights of control plants after treatment applications ($p<0.001$, ANOVA). Heignt growth in 1987 was very similar to that noted in 1986 and was significantly lower than control heights from day 67 to the end of the sampling period ($p<0.001$, ANOVA). These data suggest that there was virtually no recovery of height growth in the high marsh zone one year after the oil treatment. On the first sampling date in 1987 (Day 26), oiled plants were significantly taller than control plants. This trend was observed in all vegetation zones although it was significant only in the creek edge and high marsh zones. Reductions in stem density associated with the oil treatments may have reduced competition allowing more rapid growth early in the growing season.

Stem Density

Stem density data for the creek edge zone in 1986 and 1987 are presented in Figure 10. The oil treatment was associated with significant reductions in stem density on all sampling dates following treatment application (Appendix 4). Data from 1987 demonstrate an increase in stem density. No

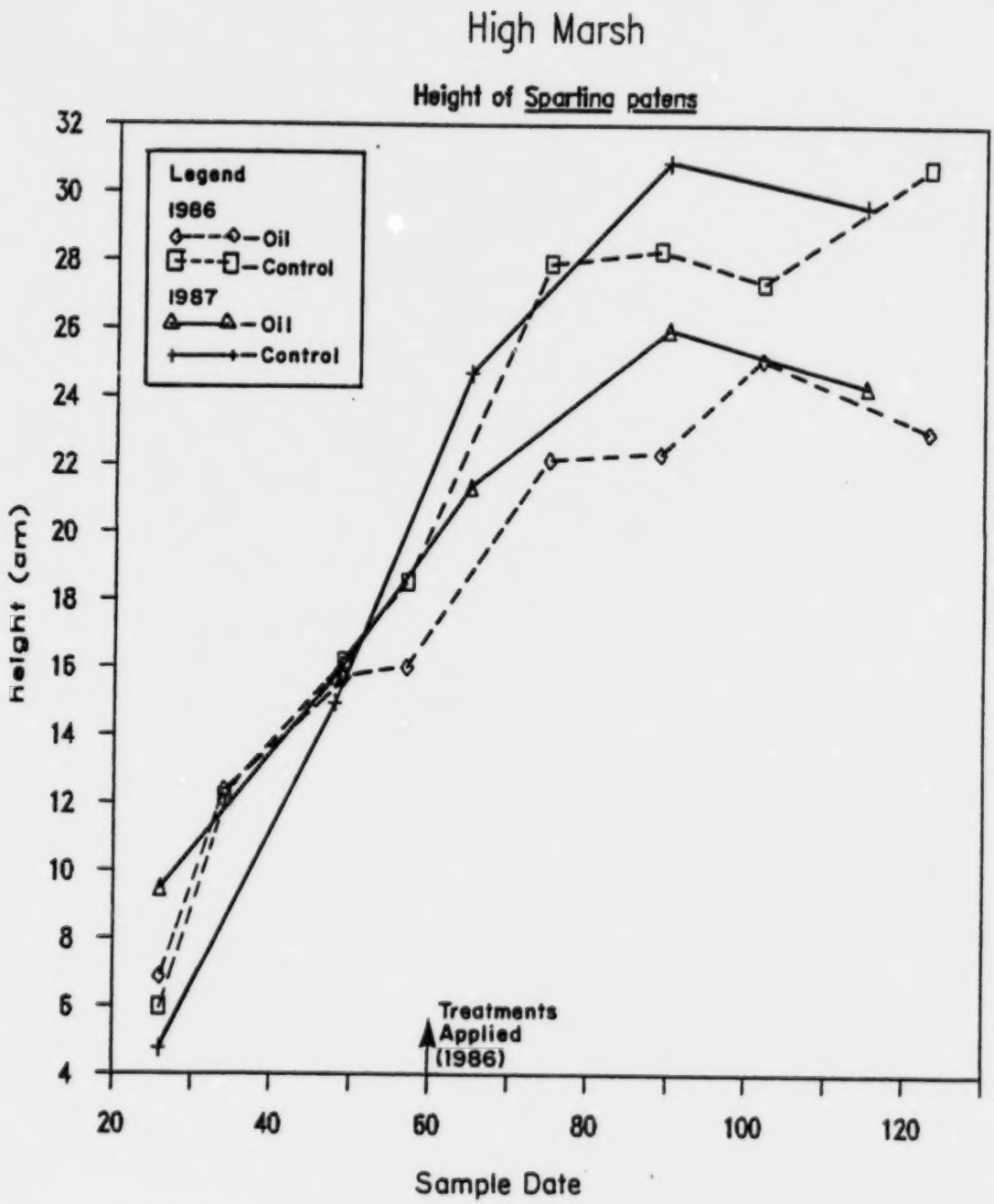


Figure 9: Plot of mean Spartina patens height for the high marsh zone versus time.

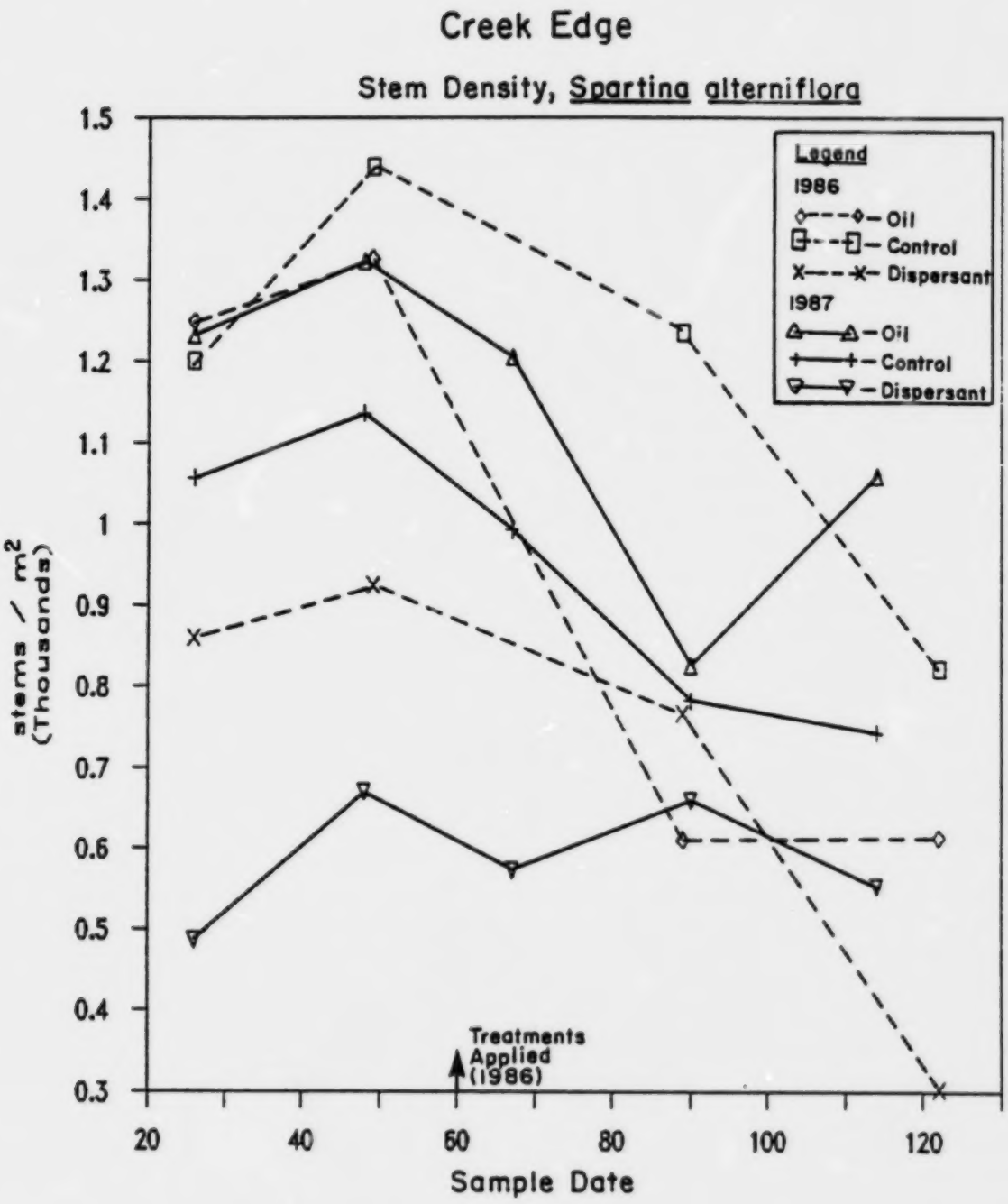


Figure 10: Plot of mean Spartina alterniflora stem density for the creek edge zone versus time.

significant differences were found between control and oiled plots in 1987 (Appendix 4). On all dates oiled plots had higher stem densities than control plots. This situation appeared to be attributable more to large reductions in control stem density rather than to substantial improvements in oiled plot stem density. Nevertheless, the data does indicate a recovery of stem density in oiled plots. No clear reasons for reductions in control stem density were evident. Small reductions in stem density were also noted in all high marsh control quadrats and two out of three midmarsh control quadrats. Trampling damage to the periphery of the quadrats may have stressed plants within the quadrats resulting in reduced stem density or yearly variations in growing conditions might also be responsible. Summer rainfall was very heavy in 1986 and light in 1987. Drought conditions might be expected to increase osmotic stress in salt marsh plants resulting in stem density reductions.

The stem density of dispersant treated plots was significantly lower than control plots on all sampling dates in 1986 including pretreatment dates ($p<0.001$, ANOVA). This indicated that control and dispersant treated plots were not directly comparable. Determination of dispersant impacts, therefore, was best determined by comparing data from 1986 and 1987 for the dispersant treated plots and comparing stem density trends for the control and dispersant treated plots. In 1986, stem density was reduced to a mean of 300 stems $/m^2$ by the end of the growing season (Day 123) from a maximum of 930 stems$/m^2$ (day 49). During 1987 stem density remained relatively stable during the growing season with 550 stems$/m^2$ at the end of the growing season (Day 118) and a maximum of 670 stems$/m^2$ (Day 48). This data suggests a moderate recovery of stem density during the 1987 growing season. At the current rate of recovery stem density in the dispersant treated plots should return to pretreatment levels within two years of dispersant applications. Stem density in control plots in both 1986 and 1987 declined gradually from a peak early in the growing season (Day 48 or 49) to minimum density at the end of the growing season (Day 115 or 123). This decline was probably attributable to mortality of shoots induced by intraspecific competition. In dispersant treated plots in 1987 stem density remained stable and plants were generally well spaced in the quadrats. This suggests that intraspecific competition was probably reduced in these quadrats resulting in less mortality. On the last two sampling dates in 1987, the stem densities of dispersant treated plots were not significantly different from control stem densities (Appendix 4).

In the midmarsh zone (Figure 11) the oil treatment was associated with significant reductions of stem density ($p<0.001$, ANOVA (89% reduction in stem density by the end of the growing season)). Stem densities of oil treated plots remained significantly lower than controls throughout the

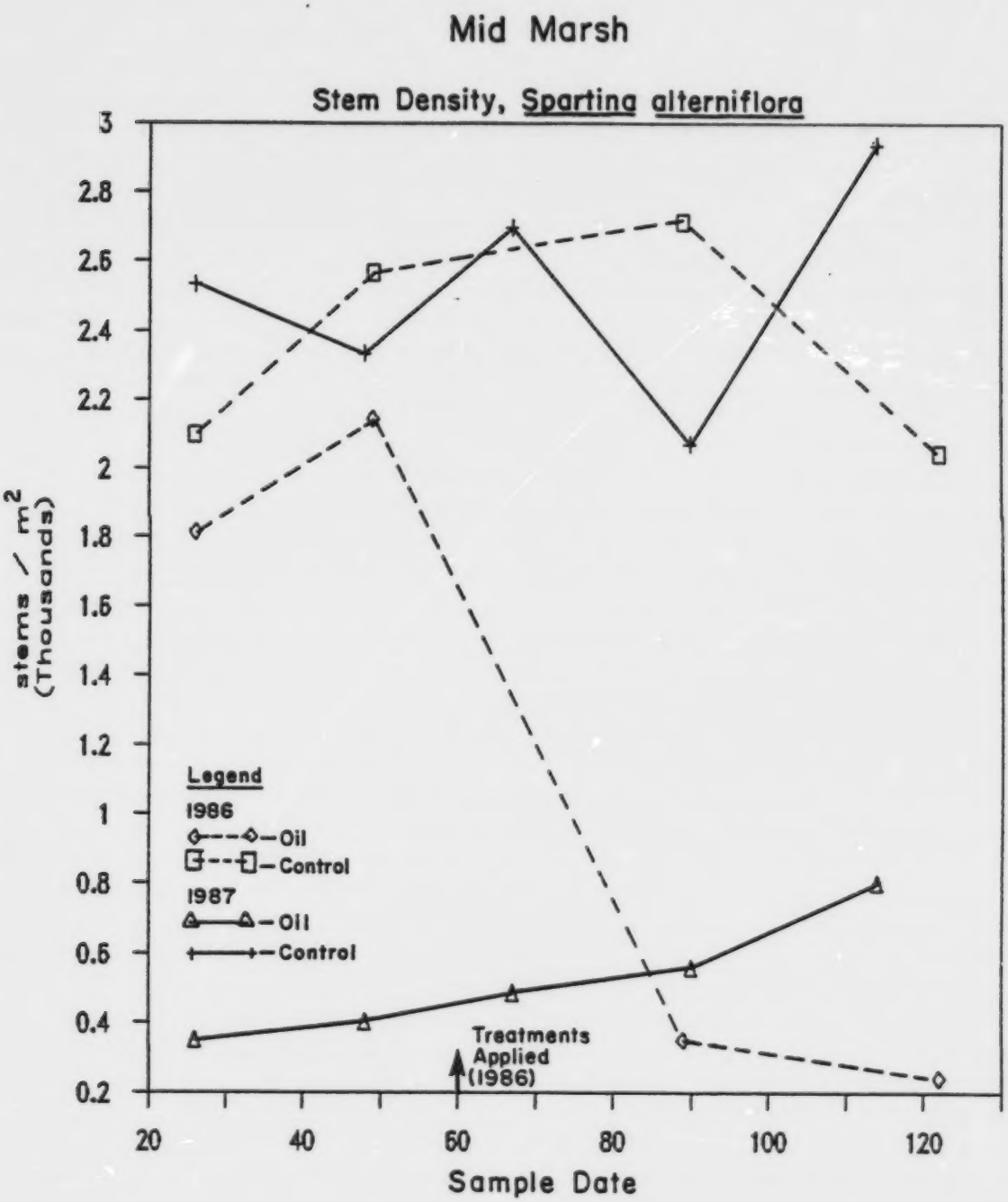


Figure 11: Plot of mean Spartina alterniflora stem density for the midmarsh zone versus time.

1987 growing season ($p<0.001$, ANOVA). Stem density early in the 1987 growing season (day 26) was 350 stems/m^2 which was similar to that noted at the end of the 1986 growing season (230 stems/m^2). Over the course of the 1987 growing season Spartina alterniflora stem density increased slowly to 800 stems/m^2. This appears to indicate that stem density recovered slightly during the 1987 growing season, however, at the end of the sampling period, the mean stem density of the control plots was still 73% higher than that of oiled plots. Complete revegetation of the midmarsh zone will probably take at least five years assuming that the current recovery rate is maintained.

Oil treatments in the high marsh zone (Figure 12) were associated with significant reductions in stem density in 1986. In 1987, oil treated quadrats had significantly lower mean stem densities on all but the first sampling date. Some recovery of stem density was evident in the data. At the end of the 1986 growing season mean stem density of oil treated plots was 48% lower than that of control plots. Stem density of oiled plots at the end of the 1987 growing season was only 32% lower than that of control plots indicating that some recovery had occurred. Complete revegetation of the high marsh might be expected to occur within three to four years of treatment application assuming the current rates of recovery are maintained.

The stem density of flowering Spartina plants varied greatly from year to year (Figure 13), however, within years the trends for the various treatments were consistent. In 1986, oil or dispersant treatments caused large declines in flowering shoot density although none of the reductions were significant (Appendix 5). For the oil treatments, the largest reduction in density was noted in the creek edge zone (99% reduction) and the smallest in the high marsh zone (64% reduction). Flowering shoot stem density in the midmarsh zone was reduced by 86%. Dispersant treated plots in the creek edge zone exhibited a 95% reduction in density. Similarly, Baker (1971) noted that oiling of Spartina x townsendii in the early summer resulted in reduced flowering. This was attributed to oil induced mortality of flower buds or primordia. Alternatively, Jefferies and Perkins (1977), suggest that under environmental conditions which require large allocations of nitrogen and carbon for maintenance (as would be expected as a result of oil toxicity) sexual reproduction may fail to occur or the development of seeds may be aborted. In 1987, the trend for flowering stem density in 1986 was reversed. Flowering shoot densities for oil and dispersant treated plots in 1987 were equal to or in most instances, higher than control densities from 1986 or 1987. In the creek edge zone, dispersant treated plots had the highest density of flowering shoots, followed by oiled plots then control plots at a ratio of 14:6:1.

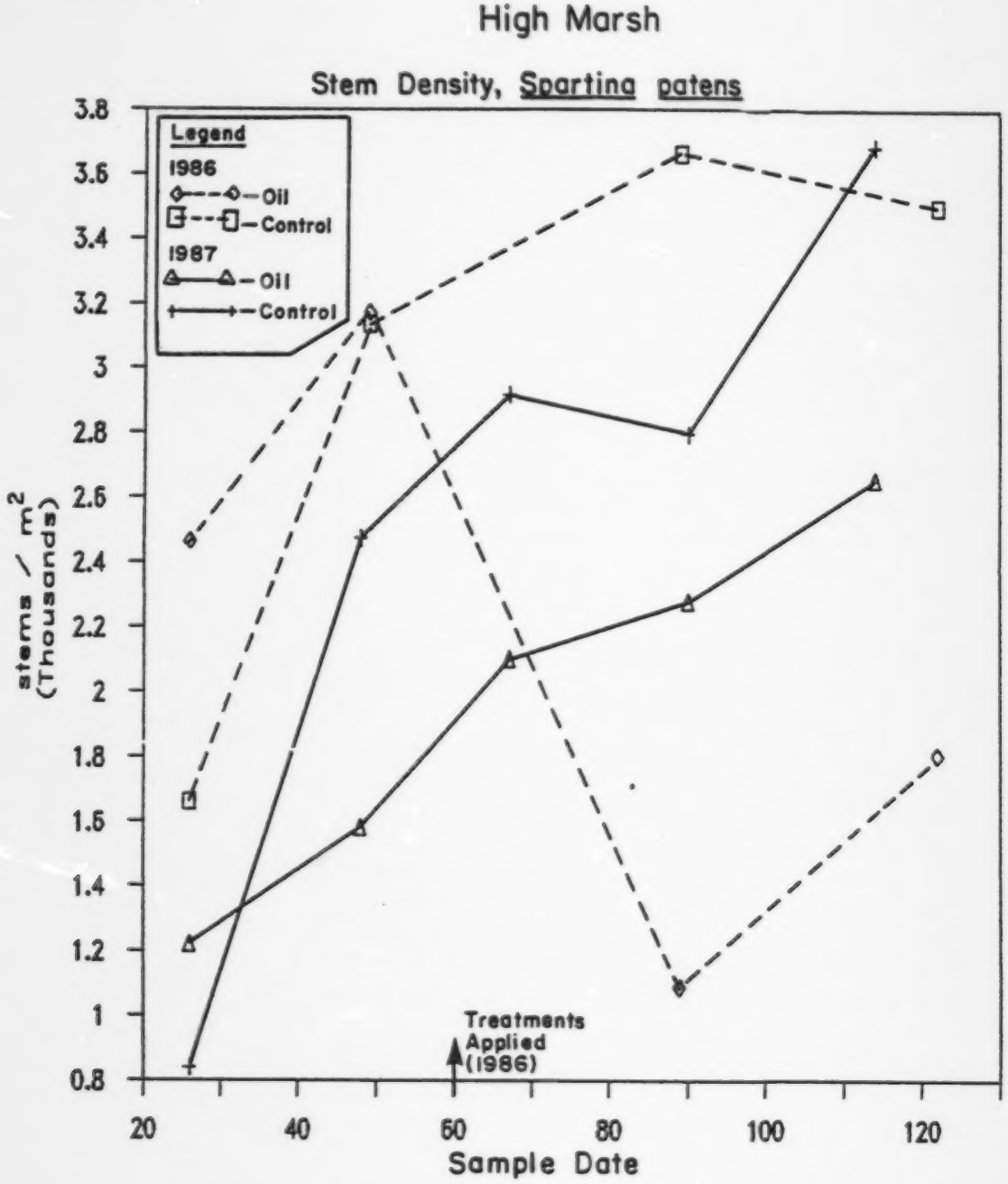


Figure 12: Plot of mean Spartina patens stem density for the high marsh zone versus time.

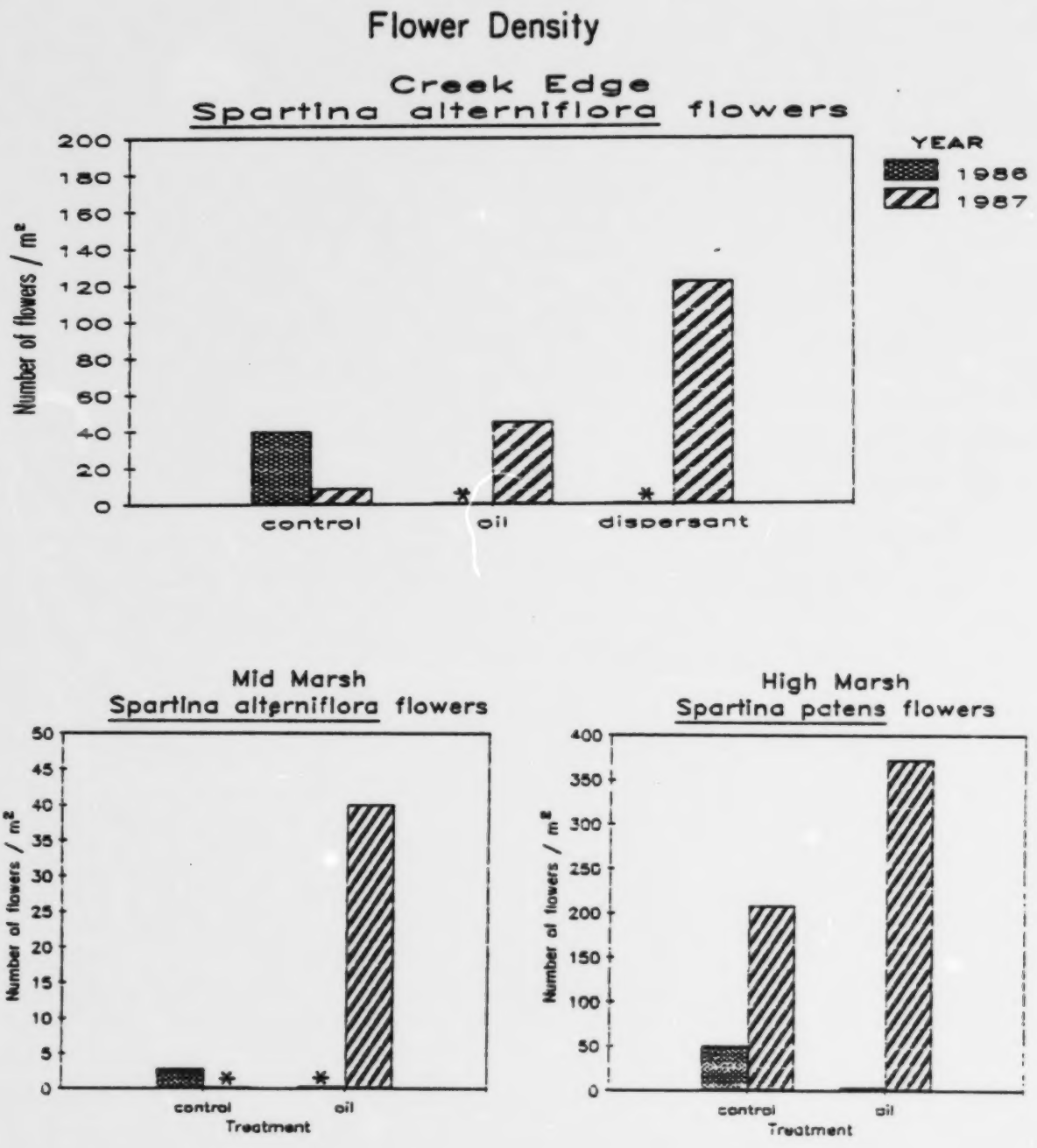


Figure 13: Histograms of mean flowering shoot density in the creek edge, midmarsh and high marsh zones.

Flowering shoot densities were significantly higher in the dispersant treated plots (p=0.004, ANOVA) but not in the oiled plots (Appendix 5). In the midmarsh zone oiled plots had 45 times more flowering shoots than control plots, however, this difference was not significant. Flowering shoot density in the high marsh zone was 1.5 times higher in the oiled plots than in the control plots but was not significantly different from the control plots. In all vegetation zones, oil treatments induced considerable, although not statistically significant, increases in flowering stem density in 1987. Lack of significance was attributable to the low number of replicates and variability of replicates within treatments. Increases in flowering shoot density appeared to be related to reductions in stem densities noted for the oil and dispersants in 1986. Stem density reductions would decrease the degree of intraspecific competition allowing more resources to be channeled into reproductive organs. Similar increases in flowering shoot density have been noted in Spartina anglica stands in Britain following cutting or trampling damage (Hubbard, 1970).

Biomass

Total above-ground living biomass data from the creek edge zone (Figure 14), indicated that the oil treatment had no significant effect on total above-ground living biomass in either 1986 or 1987 (Appendix 6). Plots treated with dispersant had significantly less biomass in 1986 (p=0.017, ANOVA) but recovered to control levels by the end of the 1987 growing season. In the midmarsh zone in 1986 (Figure 15), total above-ground living biomass in oiled plots was reduced to 15% of control plot biomass (significant p=0.003, ANOVA). In 1987 oiled plots had recovered to 58% of control biomass values and were not significantly different from the control plots (Appendix 6). Total above-ground living biomass for oiled plots in the high marsh zone (Figure 16) was reduced to 69% of control values in 1986. This reduction was not significant. Total above-ground living biomass in the oiled plots increased in 1987, however, control values also increased so that the relative amount of biomass in the oiled plots remained stable at 69% of control plot values. The variance of both control and oil treated plots declined substantially in 1987 resulting in a significant difference between the treatments (p=0.011, ANOVA).

These data suggest that the oil treatments had little impact on biomass production in the creek edge zone. In the more stressful midmarsh zone the oil treatment had severe impacts in 1986 but appeared to recover rapidly in 1987. Data from the high marsh zone suggests a moderate impact in 1986 but with relatively little recovery in 1987. The creek edge dispersant treatment was responsible for a major reduction in total above-ground

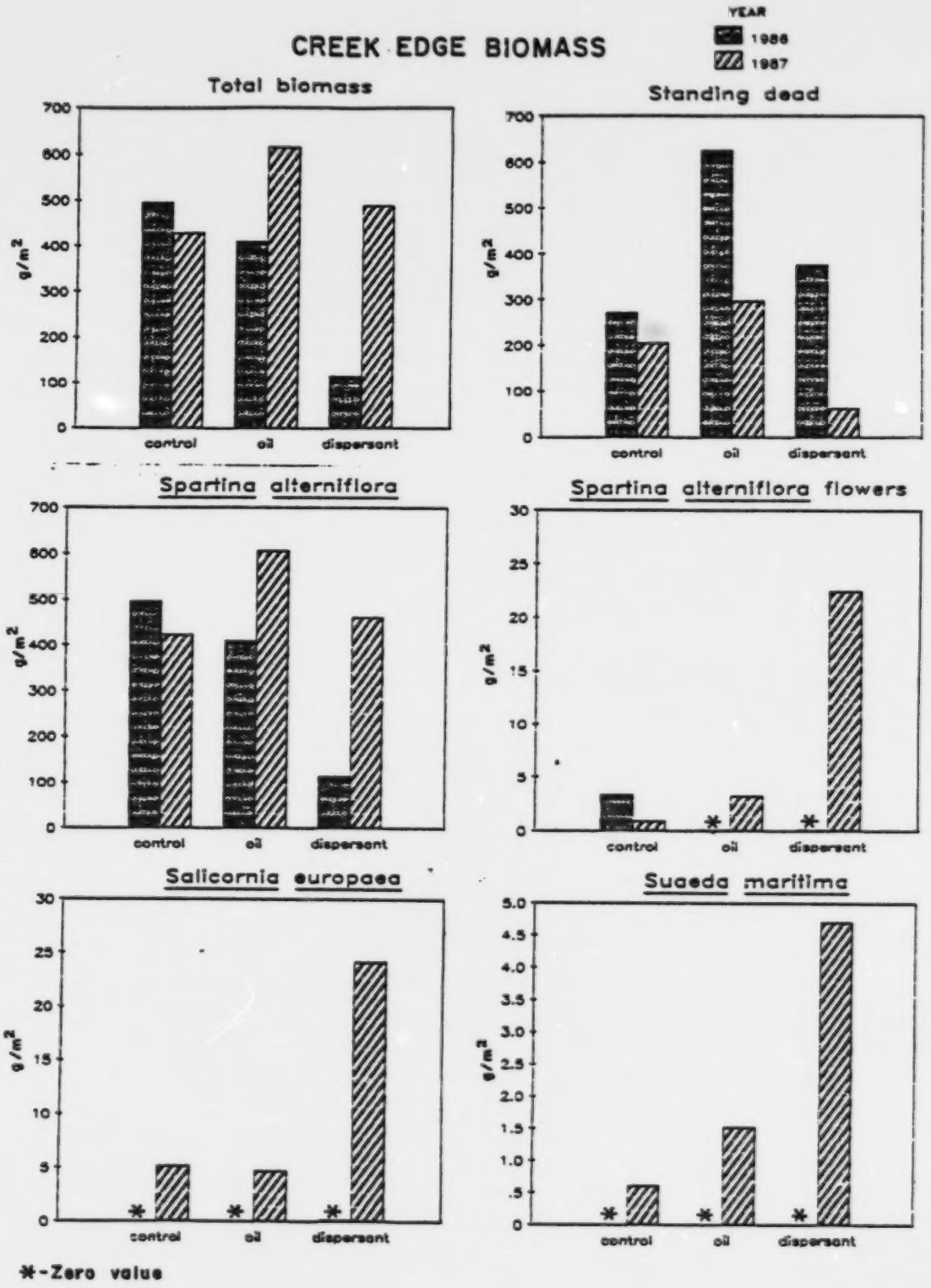


Figure 14: Histograms of mean total above-ground living biomass, standing dead biomass, Spartina alterniflora biomass, Spartina alterniflora flower biomass and Suaeda maritima biomass in the creek edge zone.

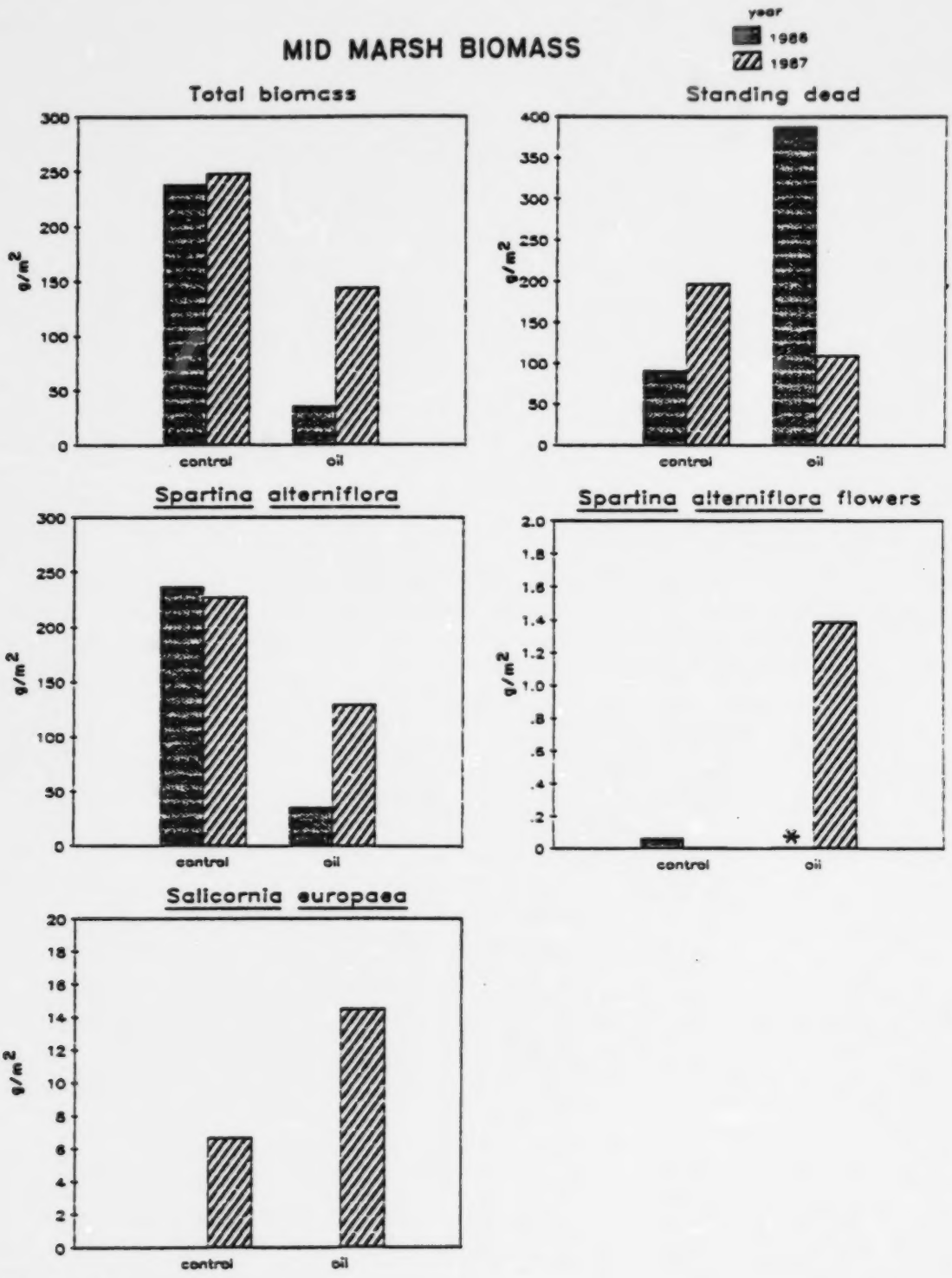


Figure 15: Histograms of mean total above-ground biomass, standing dead biomass, Spartina alterniflora biomass, Spartina alterniflora flower biomass, and Salicornia europaea biomass in the midmarsh zone.

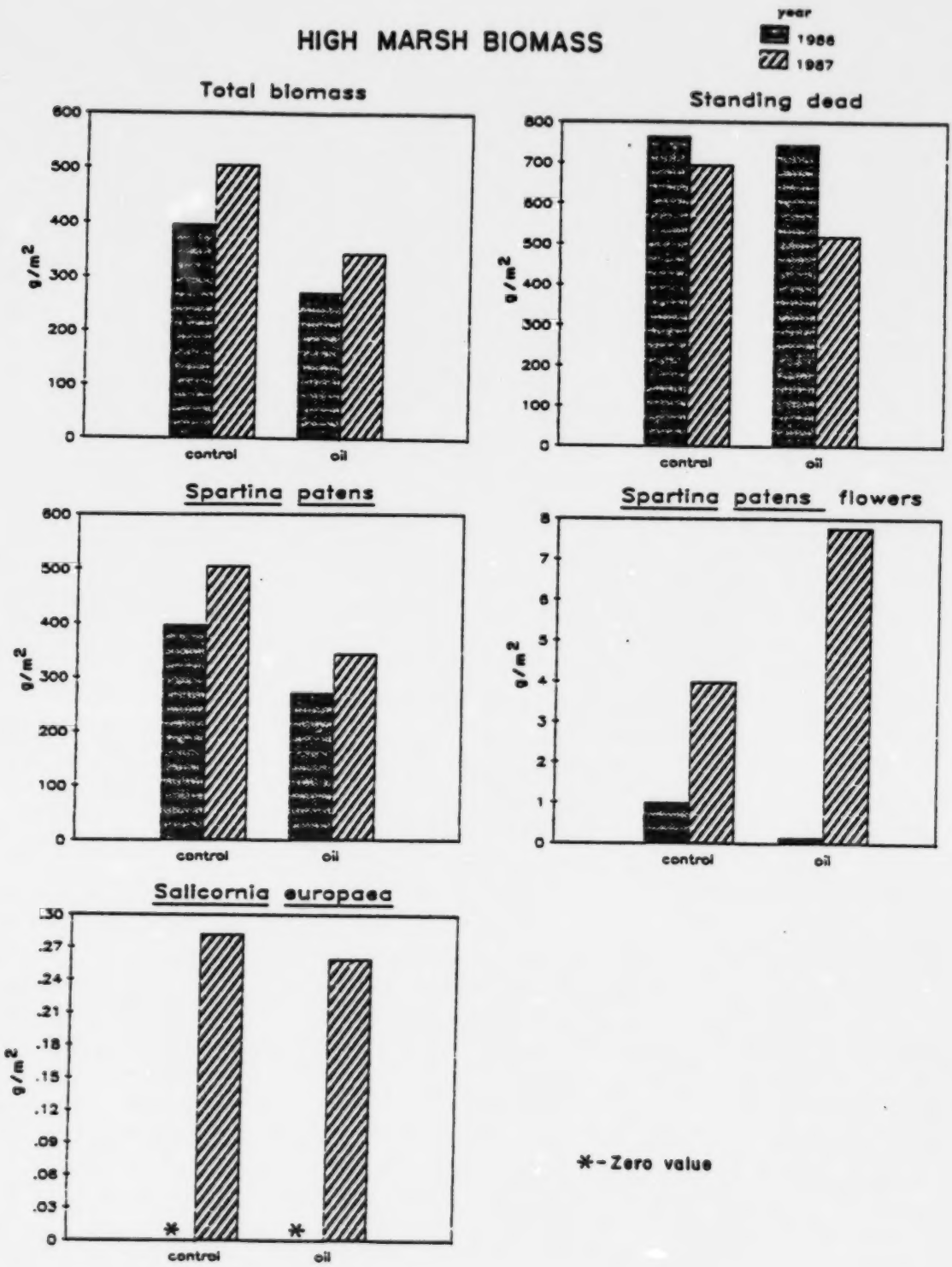


Figure 16: Histograms of mean total above-ground living biomass, standing dead biomass, Spartina patens biomass, Spartina patens flower biomass and Salicornia europaea biomass in the high marsh zone.

living biomass in 1986, however, it recovered rapidly to control levels by the end of the 1987 growing season.

Standing dead biomass in oiled creek edge plots (Figure 14) was 2.3 times higher (significant p=0.022, ANOVA) than that of the control plots in 1986. In 1987 standing dead biomass in the oiled plots was 1.5 times larger than that of the control and no longer significantly different from the controls. Standing dead biomass in dispersant treated plots was 1.4 times higher than control levels in 1986. This difference was not statistically significant. In 1987 the trend was reversed and standing dead biomass for the dispersant treated plots was only 31% of the mean value for control plots. In the midmarsh zone (Figure 15) in 1986, standing dead biomass increased by a factor of 4.2 (significant p=0.009, ANOVA) relative to control values following the application of oil. This large increase was countered by a large but insignificant reduction of standing dead biomass during the 1987 growing season. Standing dead biomass at this time was 56% of control standing dead biomass. In the high marsh zone (Figure 16) mean standing dead biomass in the oil and control plots were almost identical (no significant difference). In 1987 standing dead biomass in the oiled plots declined to 75% of control values. This decline was not significant.

Table 3 presents the proportion of total above-ground biomass (total above-ground living biomass + standing dead biomass) composed of standing dead biomass. Large increases in percent standing dead biomass were noted for the oil and dispersant treatments in the creek edge zone and the oil treatment in the midmarsh zone. This was to be expected since increased plant mortality would increase standing dead biomass. Comparison of standing dead biomass from the various treatments with plant mortality (as indicated by changes in stem density) associated with these treatments indicated that 1986 standing dead biomass values were higher than expected in the oiled plots from the creek edge and midmarsh zones. In the creek edge zone standing dead biomass was roughly equivalent to the combined total above-ground living biomass and standing dead biomass of the control plots yet stem density was reduced only 25% by the oil treatment. Similarly, standing dead biomass in oiled midmarsh plots exceeded the combined total above-ground living biomass and standing dead biomass of the controls although in this instance stem density reductions were more severe (88%). These results suggest that the oil treatment inhibited the decomposition of standing dead biomass. De la Cruz, Hackney and Rajanna (1981), have noted that oil treatments reduced decomposition of Juncus roemerianus standing dead biomass by 52%. In 1987 percent standing dead biomass for the oiled plots in all zones were identical to control values, suggesting that the rates of mortality and decomposition in the oiled plots had returned to control levels. Percent standing dead biomass for the

Table 3. Standing dead biomass (SDB), total above-ground biomass (TAB) and the percentage of total above-ground biomass composed of standing dead biomass (percent SDB) for various treatments in three vegetation zones in 1986 and 1987.

		1986			1987		
Zone	Treatment	SDB(g/m^2)	TAB (g/m^2)	Percent SDB	SDB(g/m^2)	TAB (g/m^2)	Percent SDB
Creek Edge							
	Control	272	769	35.4	206	636	32.4
	Oil	626	1040	60.4	298	913	32.7
	Dispersant	377	492	76.6	64	555	11.5
Mid Marsh							
	Control	92	330	27.8	197	446	44.3
	Oil	388	424	91.6	111	255	43.4
High Marsh							
	Control	765	1160	65.9	696	1200	57.9
	oil	746	1020	73.2	520	864	60.2

dispersant treated creek edge plots was much lower than that of the control plots in 1987. This was to be expected since total aboveground living biomass increased by a factor of 4 between 1986 and 1987. Stem density data for these plots indicated very little mortality during the 1987 growing season, therefore, inputs into the standing dead biomass would have been very low.

Trends for Spartina alterniflora biomass in the creek edge and midmarsh zones and S. patens in the high marsh zone (Figures 14 - 16) were identical to those of total above-ground living biomass in these zones since these species constituted 90 to 100% of the total above-ground living biomass. The reader is, therefore, referred to the text regarding total above-ground living biomass for details regarding the dynamics of these species.

Two annual species, Salicornia europaea and Suaeda maritima were the only other species which occurred regularly during the biomass sampling. Neither species was very common in 1986 in any of the vegetation zones (Figures 14 - 16). In 1987 biomass values for both species increased substantially although not significantly (Appendix 6) in all treatments and in all zones with the exception of the high marsh zone where small increases in Salicornia were noted and Suaeda was absent in both 1986 and 1987.

Salicornia and Suaeda typically colonize areas of bare sediment following disturbance (Ranwell, 1972; Hampson and Moul, 1978; Bertness and Ellison, 1987). In this study, Salicornia and Suaeda plants tended to be more numerous and larger in areas where the Spartina canopy had been opened up by oil or dispersant induced mortality. In control plots these species tended to persist as small scattered individuals.

In the creek edge zone (Figure 14) Salicornia biomass increased by the same amount in the control and oil plots between 1986 and 1987. There was little mortality of Spartina in the oiled creek edge plots, therefore, there was relatively little open space in which Salicornia could become established. In dispersant treated plots, where Spartina mortality was high, Salicornia biomass was 4.7 times higher than in control plots although high variability of the data rendered this difference insignificant. In the midmarsh zone (Figure 15) a substantial increase in Salicornia biomass was noted in both the control and oil treated plots between 1986 and 1987. Salicornia biomass was 2.2 times higher in the oiled plots (high Spartina mortality) than in the control plots, however, this difference was not statistically significant.

Suaeda biomass in the creek edge zone (Figure 14) increased in all treatments in 1987. The largest increase was noted in the dispersant treated plots (7.6 times higher than controls) followed by the oil treated plots (2.5 times higher than controls), however, these differences were not statistically significant.

The relatively large increases in Salicornia and Suaeda biomass noted in the creek edge and midmarsh control plots between 1986 and 1987, were probably related to reductions in Spartina alterniflora biomass in these plots. Reductions of S. alterniflora biomass were probably attributable to a combination of drought conditions in 1987 and sampling disturbance during both years. Salicornia is very tolerant of high salinities (Brereton, 1971). Hypersaline conditions experienced during the very dry 1987 growing season may have reduced Spartina density allowing the more salt tolerant Salicornia to proliferate.

Reproductive biomass varied considerably between 1986 and 1987 (Figures 14 - 16). In the creek edge and midmarsh zones Spartina alterniflora flower biomass in control plots declined in 1987 while in the high marsh zone S. patens flower biomass increased. In the midmarsh and high marsh zones these differences between years were significant (midmarsh $p=0.020$, high marsh $p<0.001$, ANOVA). There are no clear reasons for these declines. Drought conditions during the 1987 growing season may have reduced the incidence of flowering. Within years distinctive trends were noted. In 1986 oil and dispersant treatments caused large reductions in flower biomass in all zones. These reductions, however, were not significant as a result of high variability of the data and the low number of replicates. In 1987, the opposite trend was noted. Increases in flower biomass were noted in treated plots. Increases associated with the oil treatments were not significant as a result of low replicate number and high variability of the data. The dispersant treatment in the creek edge zone was associated with a very large increase in flower biomass which was significant ($p=0.006$, ANOVA). These increases appeared to be related to the intensity of disturbance. Plots in which mortality was high in 1986 and had displayed vigorous recovery in 1987 tended to have the highest flower biomass. Reduction of stem density would decrease intraspecific competition, thereby, increasing the availability of resources required for flowering.

No significant differences in mean plant weight were found between any of the treatments in the creek edge, midmarsh on high marsh zones in either 1986 or 1987 (Appendix 6), however, some general trends were observed in the data. In the creek edge zone mean plant weights for control and oiled plants were similar in both 1986 and 1987 suggesting no short term or long

term effects of the oil treatment on plant growth (Figure 17). A reduction in mean plant weight was noted in both the control and oiled plants between 1986 and 1987, possibly as a result of drought conditions during the 1987 growing season. Dispersant treated plants averaged half as heavy as control plants in 1986 indicating a substantial reduction in growth. In 1987 dispersant treated plants were 1.4 times heavier than control plants. Stem density was low and plants were well spaced in the dispersant treated plots. Interspecific competition would have been low in these plots allowing greater growth. These data also suggest that the toxic effects of the dispersant had probably disappeared by 1987. This would be expected since the dispersant is miscible in water and would be easily flushed from the plots.

In the midmarsh zone oiled plants were 1.3 times heavier than control plants in 1986. This was probably caused by selective mortality of short plants. Short plants would have a larger proportion of their surface area covered by the oil and would be expected to suffer higher mortality. In 1987 the oiled plants were 2.0 times larger than the controls. Reductions of stem densities in the plots may have alleviated intraspecific competition allowing greater growth of plants in the oiled plots, however, plant height data indicated that these plants were significantly shorter than controls and fluorometry data indicated some residual oil toxicity. This would indicate that large changes in plant morphology had occurred, specifically that small but robust tillers were produced in response to the oil treatment. Hershner and Lake (1980) also found changes in plant morphology in response to oiling. Contrary to our findings, they observed that oiled plants were generally the same height as control plants but more slender. There was a marked reduction in the mean weight of control plants between 1986 and 1987 as was observed in the creek edge zone. This was probably attributable to drought conditions in 1987.

In the high marsh zone the mean weight of oil treated plants was 1.5 times higher than that of control plants in 1986. This increase was probably caused by selective mortality of small plants in 1986. In 1987 the mean weights of control and oiled plants were similar indicating the oil had little effect on plant growth. Between year differences in the controls were small suggesting that drought conditions had little effect on this species.

MEAN SHOOT WEIGHT

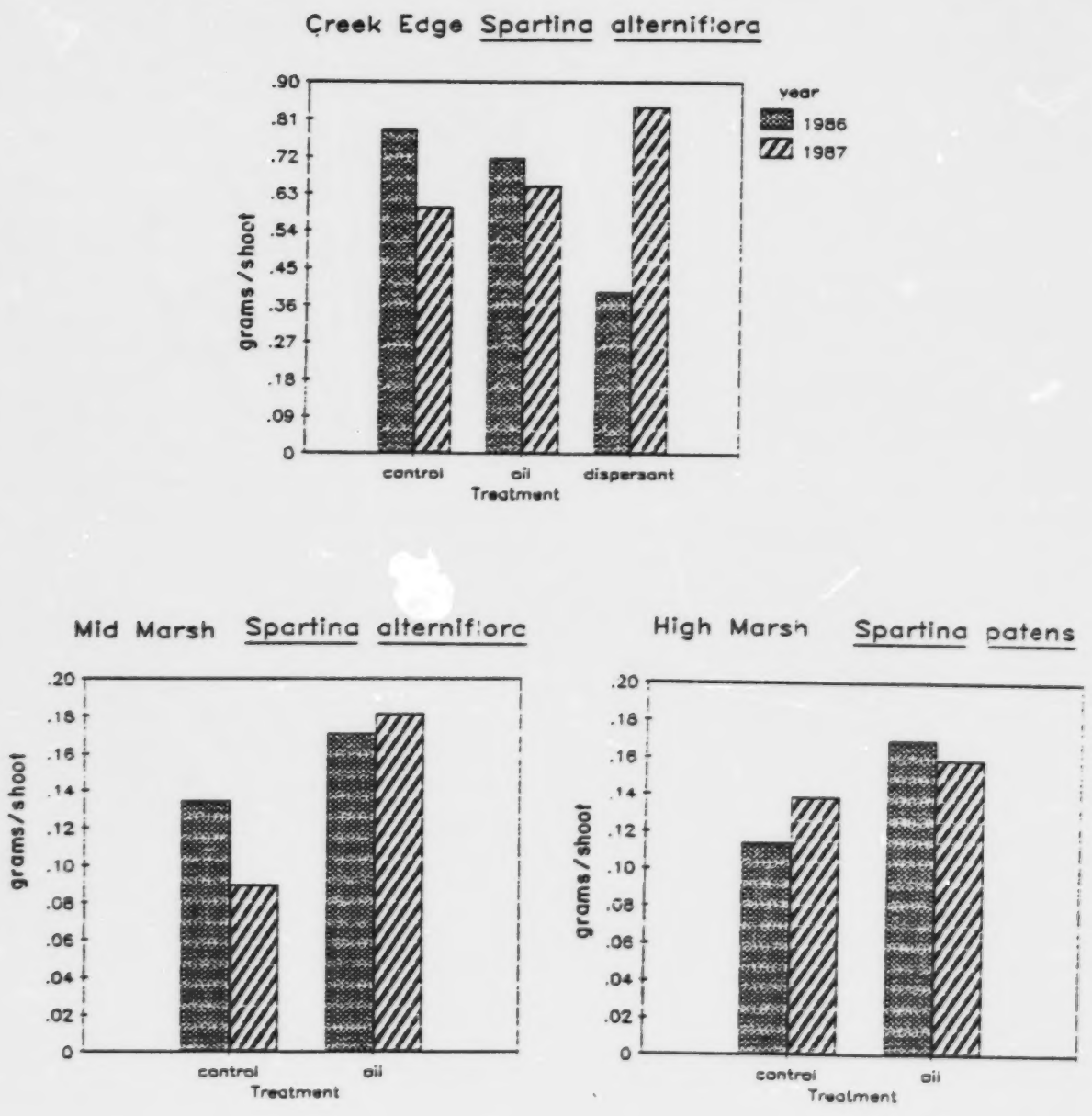


Figure 17: Histograms of mean weight of individual Spartina snoots in the creek edge, midmarsh and high marsh zones.

Species Cover

Spartina alterniflora cover in the creek edge zone was significantly reduced by the oil treatment in 1986 (Figure 18; Appendix 7). In 1987, S. alterniflora cover in the oiled plots was not significantly different from the control values, however, this apparent recovery was attributable more to a reduction in control cover in 1987 than to an increase in the cover of oiled plots in 1987. A similar trend was noted for S. alterniflora cover in dispersant treated plots. Cover reductions were significant throughout the 1986 growing season. Cover values noted during the 1987 growing season were similar to those recorded in 1986, however, on days 70 and 90 these values were not significantly different from the 1987 control values as a result of the reduction of control cover values in 1987. There is no clear reason why control cover declined in 1987. Drought conditions during the 1987 growing season may have contributed to this decline. If drought was a factor affecting the growth of controls, it would also be expected to affect growth in the treated plots. The apparent lack of recovery in these plots may be the result of an overall growth reduction caused by the drought.

In the midmarsh zone in 1986 the oil treatment was associated with significant reductions in the cover of Spartina alterniflora (Figure 19) on days 75 ($p<0.001$, ANOVA) and 102 ($p<0.001$, ANOVA). Mortality tended to occur in patches with areas of complete mortality bordering areas where mortality was very light. During the 1987 growing season no apparent expansion of surviving Spartina patches was noted, nor were new patches established, consequently, the cover noted at the end of the 1987 growing season was similar to that noted at the end of the 1986 growing season. A small increase in stem density was noted during the 1987 growing season (Figure 11). This increase was probably restricted to the interior and periphery of existing patches accounting for the lack of patch expansion noted in the cover data. During the 1987 growing season Spartina alterniflora cover in the oiled plots was significantly lower than in control plots on all sampling dates ($p<0.001$, ANOVA). The large reduction of S. alterniflora cover in the midmarsh zone relative to the creek edge zone was probably attributable to the higher physiological stress levels encountered in the midmarsh zone. S. alterniflora in this zone was unable to tolerate the additional stress associated with the oil treatment.

In the high marsh zone Spartina patens cover in the control and oil treated plots displayed little change between 1986 and 1987 (Figure 20). In 1986, the oil treated plots had significantly higher cover than control plots on day 35 (pretreatment, $p=0.048$, ANOVA), however, within fifteen days of oil application, cover in the oiled plots had declined to 90% of control

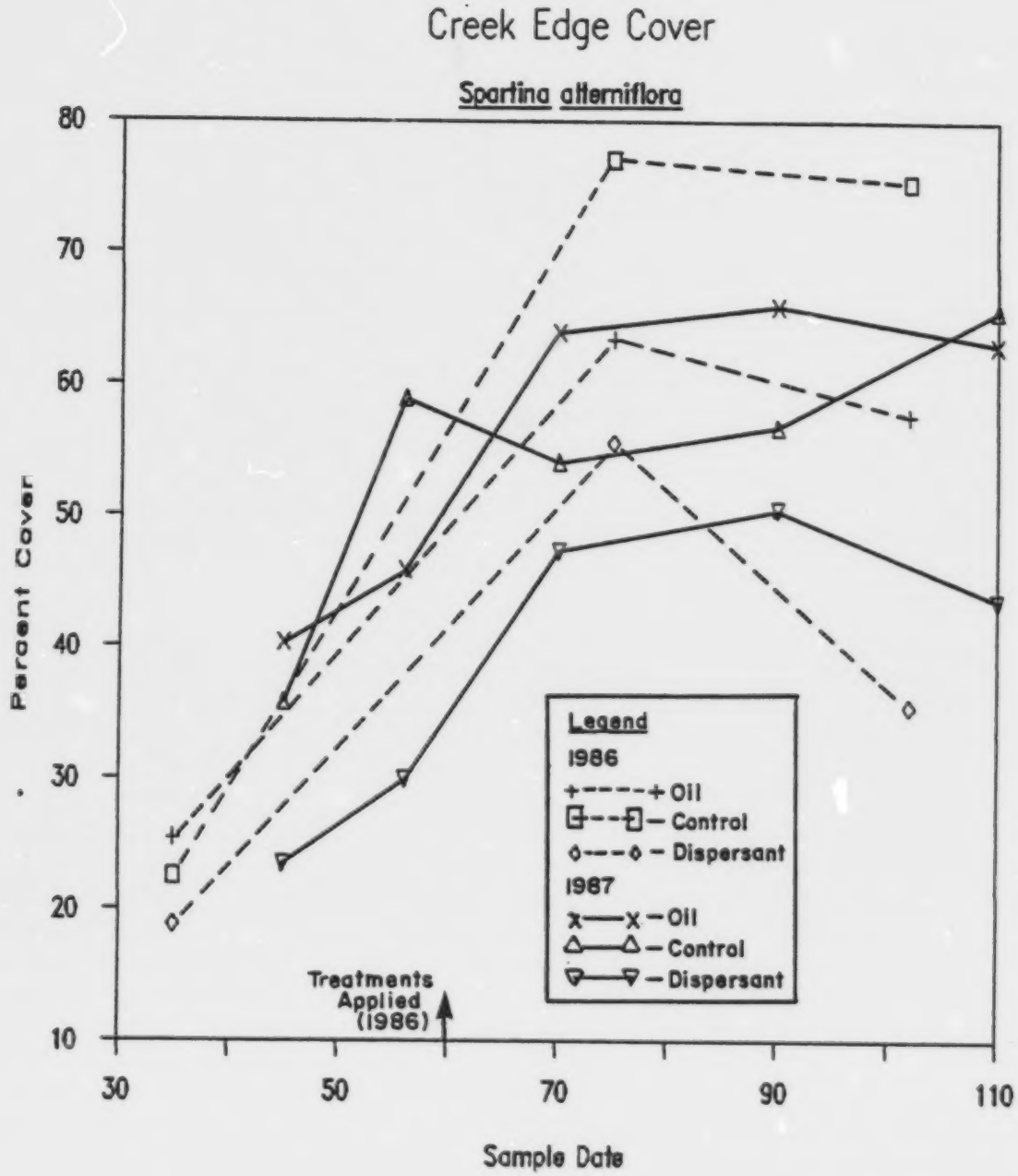


Figure 18: Plots of mean Spartina alterniflora cover in the creek edge zone versus time.

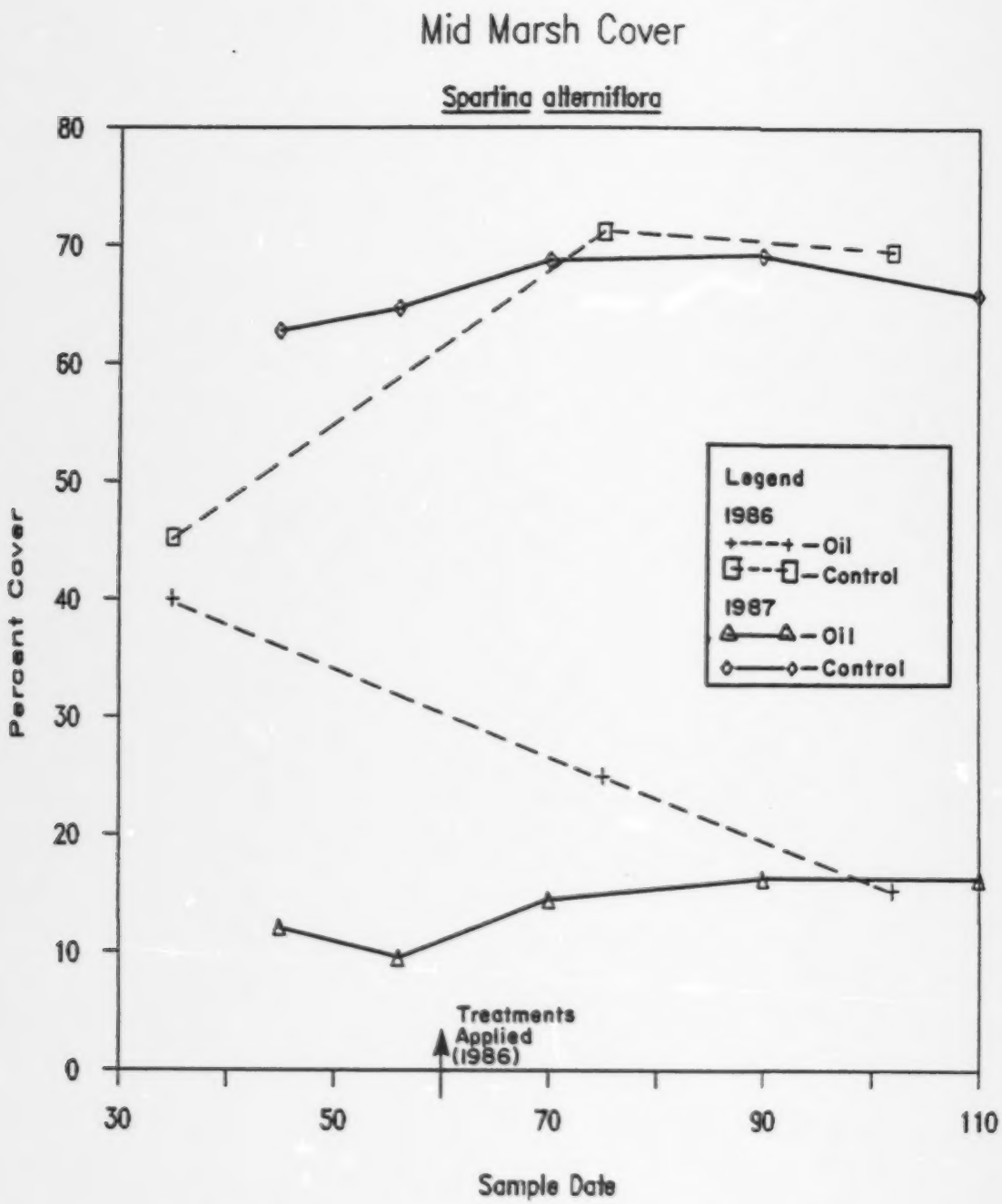


Figure 19: Plot of mean Spartina alterniflora cover in the midmarsh zone versus time.

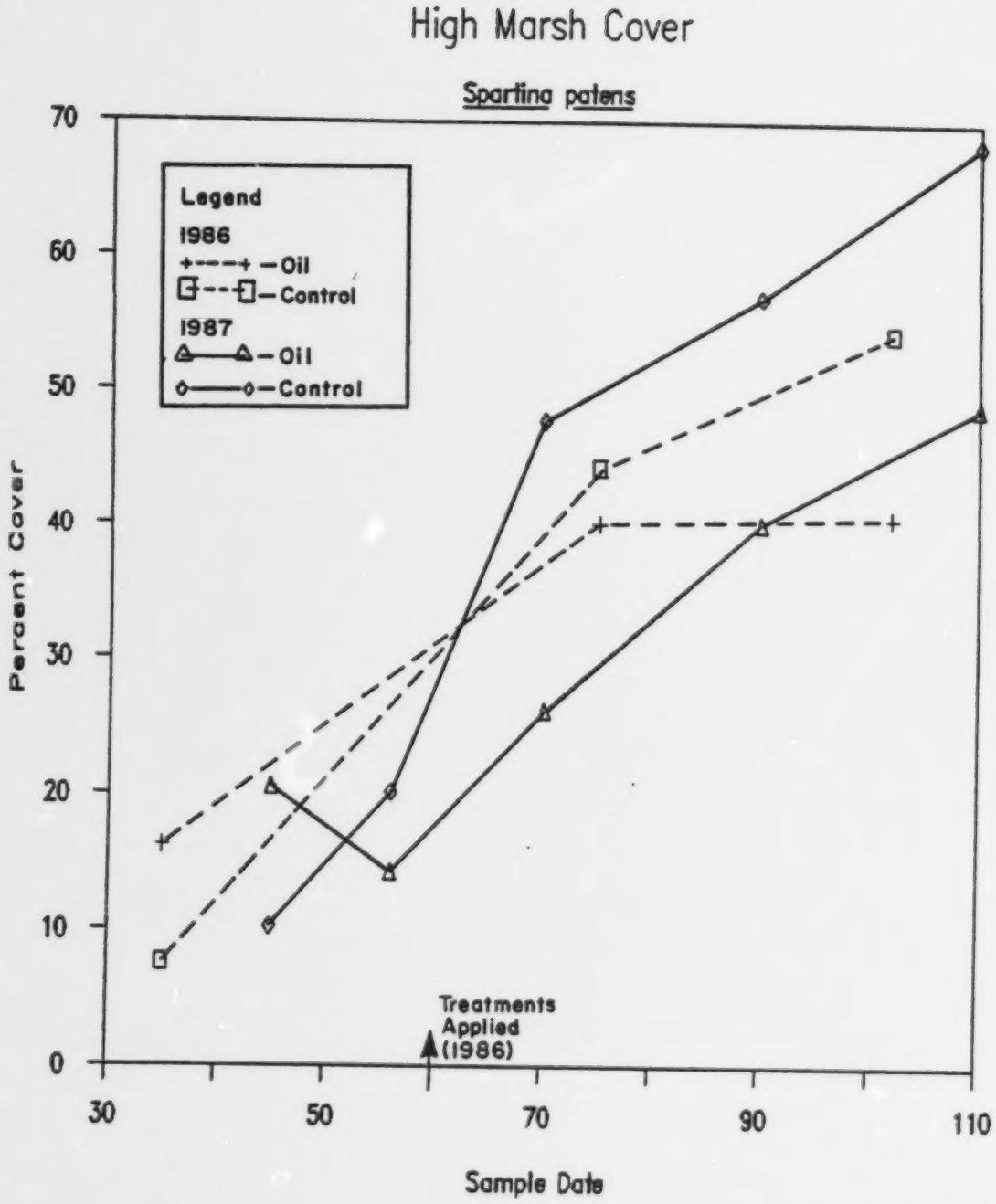


Figure 20: Plot of mean Spartina patens cover in the high marsh zone versus time.

values. S. patens cover was significantly lower than control values (p=0.005, ANOVA) on day 102. A similar trend was observed in 1987. S. patens cover in the oiled plots was significantly higher than in control plots (p=0.011, ANOVA) at the beginning of the growing season. It declined briefly on day 56 but recovered to 1986 oiled plot values by day 90. S. patens cover in oiled plots was significantly lower than in control plots on days 70 (p=0.001, ANOVA) and 110 (p=0.031, ANOVA).

This evidence suggests that there was no recovery of S. patens in the high marsh oiled plots. Reinvasion of bare patches by S. patens tillers is slow (Bertness and Ellison, 1987) and this species generally has very poor seed set as a result of seed predation by insects (Bertness et al., 1987). S. patens stands also maintain high detritus cover (up to twice the cover of living plants) which can interfere with the recolonization of bare patches by shading of seedlings. Removal of this detritus has been demonstrated to increase stem density by a factor of 4 (Lane et al., 1987). The detritus layer in the oiled plots remained intact throughout 1986 and 1987.

Salicornia europaea and Suaeda maritima were the most common species in the creek edge study plots other than the two Spartina species. Generally, they accounted for less than 10% of the total cover of the plots. During the 1986 growing season these annual species were uncommon in the creek edge zone (Figure 21). In 1986, Salicornia was eliminated by the oil treatment but appeared to be unaffected by the dispersant treatment. Cover values remained low and variable throughout the 1986 growing season, consequently none of these changes were statistically significant. In 1987, Salicornia cover increased in all treatments. Cover in control plots in 1987 was on average twice as high as control cover in 1986. This increase may have been attributable to sampling disturbance at the edges of the plots. It may also have been related to the drought induced reductions in Spartina cover mentioned above. Salicornia is an early successional species which is very tolerant of high salinity (Brereton, 1971). Drought conditions may have increased salinity to a point where it was stressful to Spartina but within the tolerance range of Salicornia. Salicornia cover in the oil and dispersant treated plots displayed similar trends during the 1987 growing season. Cover remained stable between days 45 and 56 after which it began to increase rapidly. In the oiled plots, cover increased until day 90 after which it stabilized. End of season cover was 3.0 times higher than control values. Cover in the oiled plots was significantly higher than controls on days 70 (p=0.044, ANOVA) and 90 (p=0.023, ANOVA). In the dispersant treated plots, Salicornia cover increased until the end of the sampling period. End of season cover in the dispersant treated plots was 9.5 times higher than control plots and 3.2 times higher than oil treated plots. Cover in dispersant treated plots was significantly higher

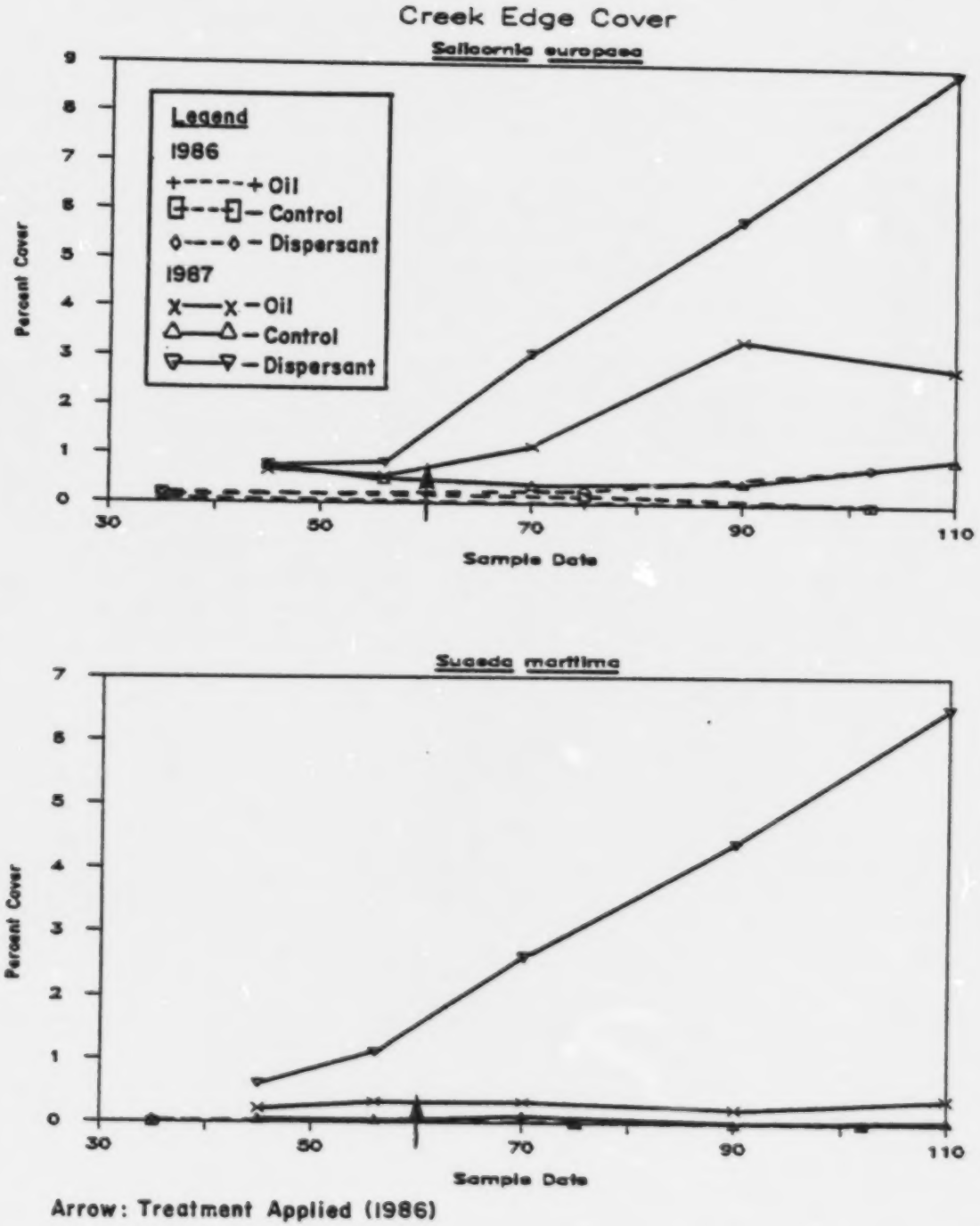


Figure 21: Plots of mean Salicornia europaea and Suaeda maritima cover in the creek edge zone versus time.

than in control plots on days 70 ($p<0.001$, ANOVA), 90 ($p<0.001$, ANOVA) and 110 ($p<0.001$, ANOVA).

The abundance of Salicornia was inversely related to the cover of Spartina alterniflora. A correlation coefficient of -0.815 was noted between the square root of Spartina alterniflora cover and the square root of Salicornia cover. Salicornia normally grows on open mudflats or in areas where stranded debris has eliminated the dominant vegetation (Bertness and Ellison, 1987). Mortality of Spartina shoots associated with the oil and dispersant treatments would open up the canopy enough to allow the establishment and maximum growth of this species. There was a curvilinear relationship between the cover of Salicornia and S. alterniflora cover. Salicornia cover was affected relatively little by reduction of S. alterniflora cover to a minimum of 50%. Below 50% cover, Salicornia cover increased rapidly.

Salicornia cover in the midmarsh zone did not respond to destruction of the Spartina alterniflora sward as vigorously as noted in the creek edge zone (Figure 22). Spartina cover in the midmarsh zone was reduced to 22% of control values by the oil treatment while the dispersant treatment in the creek edge zone reduced Spartina cover to 47% of control values, yet Salicornia cover was similar in both treatments. The strong negative correlation of Spartina cover and Salicornia cover noted in the creek edge zone was not noted in the midmarsh zone ($r=-0.656$). High physiological stress associated with the midmarsh zone accompanied by residual oil toxicity, may have contributed to the relatively poor growth of Salicornia in the midmarsh zone. This is confirmed by Brereton (1971) who found that Salicornia grew poorly in soils with low redox potential. A relatively large increase in Salicornia cover was noted in the control plots. The seasonal trend for Salicornia cover in the control plots was similar to that of the oiled plots but averaged 65% as high. This prevented the increase in cover in the oiled plots in 1987 from being statistically significant (Appendix 7). Increased Salicornia cover in the control plots was attributable to sampling disturbance which permitted the establishment of Salicornia along the edges of the plot and/or drought conditions which reduced the cover of the Spartina sward.

Salicornia cover in the high marsh zone demonstrated little yearly variation in the control plots (Figure 23). In the oiled plots the seasonal trend for Salicornia cover in 1986, was very similar to that of the control plots suggesting little oil induced mortality. In 1987, an increase in Salicornia cover was noted in the oiled plots. Salicornia cover was significantly higher in the oiled plots than in the control plots on all sampling dates ($p = 0.01$, ANOVA) except day 56. This is somewhat

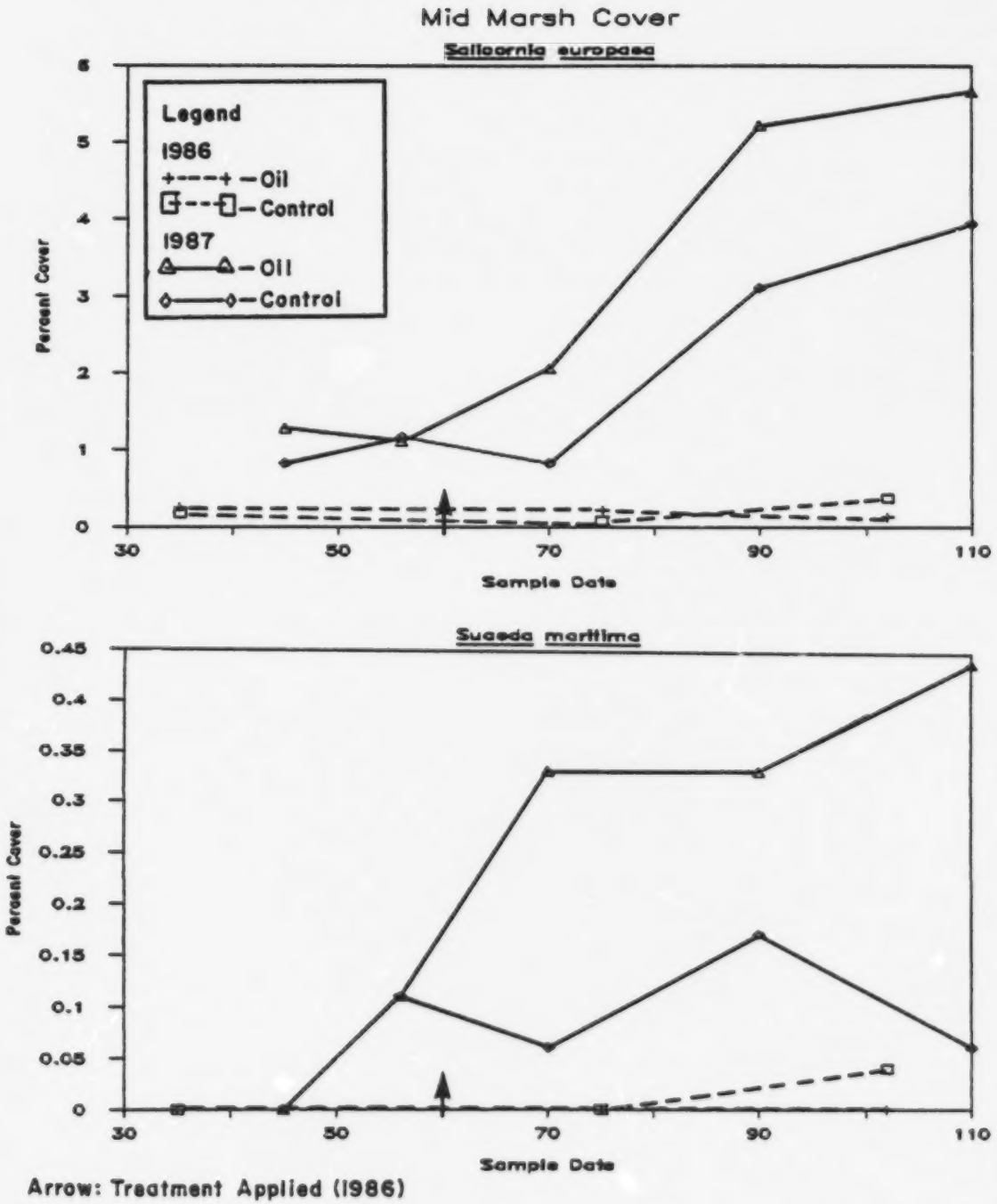


Figure 22: Plots of mean Salicornia europaea and Suaeda maritima cover in the mid marsh zone versus time.

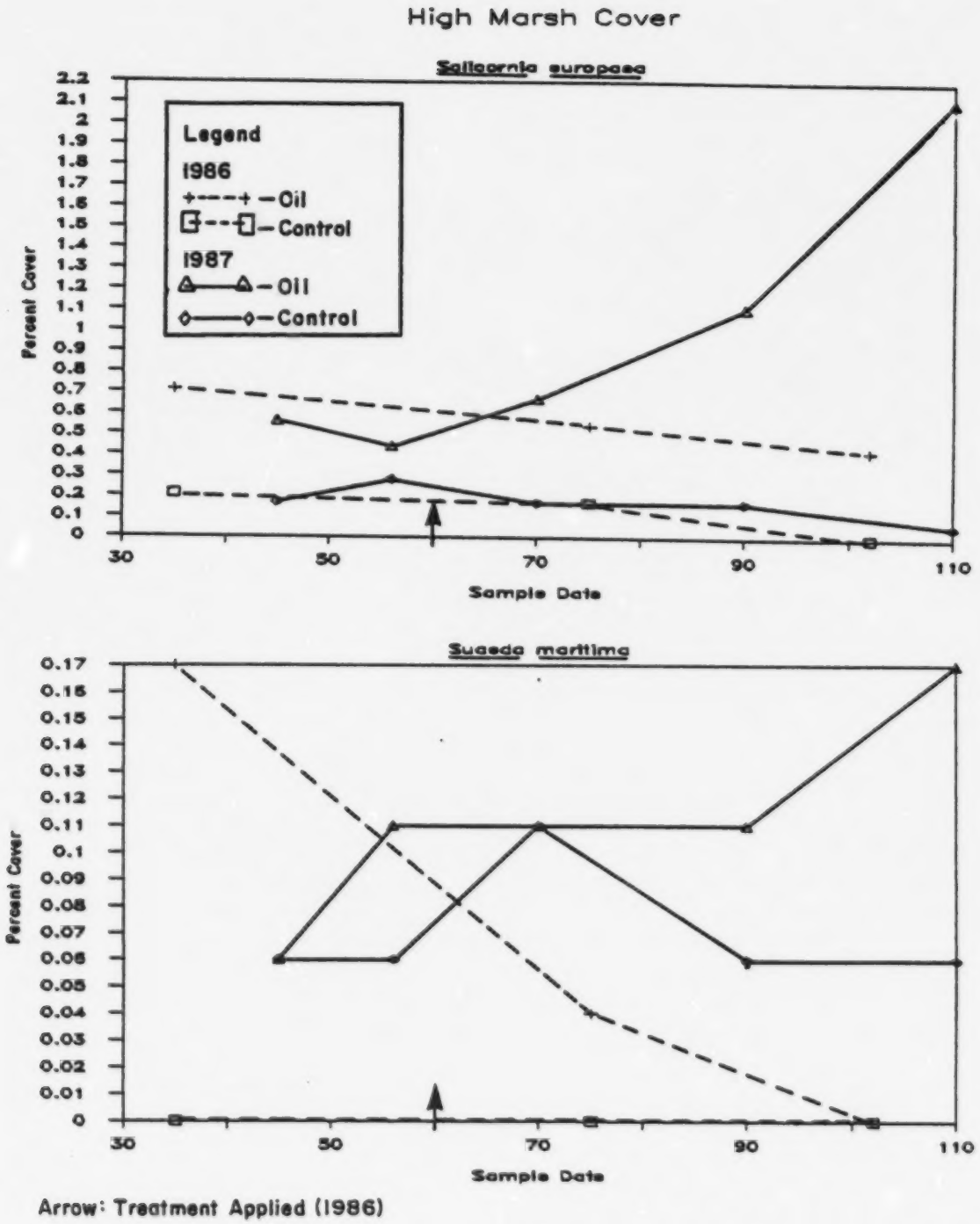


Figure 23: Plots of mean Salicornia europaea and Suaeda maritima cover in the high marsh zone versus time.

misleading since Salicornia cover was consistently higher in the oiled plots than in control plots both before and after treatment applications. In 1986, Salicornia cover in oiled plots was significantly higher than in control plots on days 35 ($p=0.014$, ANOVA) and 102 ($p=0.001$, ANOVA). There was a very strong negative relationship between Spartina patens cover and Salicornia cover ($r=-0.942$) suggesting that increases in Salicornia cover were related to reductions in S. patens competition.

Suaeda cover responded to the oil and dispersant treatments similarly to Salicornia (Figure 21). In 1986, Suaeda was present in only one dispersant plot on day 35. It disappeared from the creek edge zone following the dispersant treatment application. In 1987 Suaeda occurred sporadically in the control plots, remaining at low levels throughout the growing season. In the oiled plots Suaeda was more common, however, its cover remained stable throughout the growing season. Suaeda cover in the oiled plots was significantly higher than control plots on day 56 ($p=0.030$, ANOVA). Few Suaeda shoots grew larger than 5 cm under the well developed Spartina canopy in the oiled plots. Suaeda cover was highest in the dispersant treated plots. Cover increased throughout the growing season, reaching a peak of 6.50%. Suaeda cover in dispersant treated plots was significantly higher than that of control plots on all sampling dates in 1987 ($p<0.001$, ANOVA). There was a strong negative correlation ($r=-0.825$) between the square root of Suaeda cover and the square root of Spartina alterniflora cover suggesting that this species was responding to the removal of the dense Spartina canopy. As was noted for Salicornia, Spartina cover reductions of at least 50% were required to evoke a large increase in Suaeda cover.

In the mid marsh zone Suaeda was noted in only one control plot during the 1986 growing season (Figure 22). In 1987, Suaeda was absent in control and oiled plots on day 43 but appeared in both treatments by day 56. Suaeda cover in the oiled plots increased throughout the growing season while cover in the control plots stabilized after day 56. Suaeda cover in the oiled plots was significantly higher than in the control plots on days 70 ($p=0.044$, ANOVA) and 110 ($p=0.011$, ANOVA). Suaeda cover in the midmarsh oiled plots was equivalent to that found in oiled plots in the creek edge zone. A larger amount of cover would be expected in the midmarsh zone since Spartina mortality was much higher there. The coefficient of correlation between the square root of Spartina cover and the square root of Suaeda cover in this zone was only -0.717 as opposed to -0.825 in the creek edge zone. The combination of a normally hostile environment for plants combined with residual oil toxicity (as outlined for Salicornia) may have restricted the establishment and growth of Suaeda in the midmarsh oiled plots.

In the high marsh zone, Suaeda was present only in the oiled plots in 1986 (Figure 23). Suaeda cover in the oiled plots declined after oil application and had dissappeared from the plots by the last sampling date. This data suggests that Suaeda was sensitive to the oil treatment. Baker (1971) noted that oil treatments applied during the growing season caused severe reductions in Suaeda maritima cover. In 1987, Suaeda cover increased in both the control and oiled plots with the highest cover values noted in the oiled plots. These differences were not significant. There was a moderately strong negative relationship between Suaeda cover in the high marsh zone and Spartina patens cover (r=-0.769).

Another annual species, Atriplex patula was found sporadically in the creek edge zone in all treatments. Trends for Atriplex abundance were similar to those of Salicornia and Suaeda except that Atriplex cover declined markedly between days 90 and 100 in 1987 (Figure 24). No reason for this decline was evident.

Atriplex patula was not found in any plots in the midmarsh zone in 1986, however, it was found in some plots in 1987 (Figure 25). On days 45 and 56 Atriplex was restricted to the oiled plots, however, it became established in one of the control plots by day 70. Maximum Atriplex cover was noted between days 45 and 56 in the oiled plots. Atriplex cover in the oiled plots declined to a minimum by day 90 then increased somewhat by day 110. This cover reduction may have been caused by hypersaline conditions induced by drought conditions experienced in 1987. The increase in cover in the control plots may have been the result of increased illumination of the sediment surface caused by sampling disturbance to the edges of the control plots.

Atriplex patula cover in the high marsh zone responded similarly to Suaeda (Figure 26). In 1986 there was a decline in Atriplex cover associated with the oil treatment, however, cover returned to pretreatment levels by the end of the growing season. Increased Atriplex cover values were noted in both the control and oil treated plots in 1987 with the largest increases associated with the oiled plots. The coefficient of correlation between Spartina patens cover and Artriplex cover was -0.758.

Plantago juncoides was found in one dispersant plot in the creek edge zone. This species was absent or overlooked on the pretreatment sampling date in 1986 (Figure 24). Plantago cover increased throughout the 1986 growing season and continued to increase at a slower rate through the 1987 growing season. This data suggests that Plantago juncoides was tolerant of the dispersant application. The dominant Spartina alterniflora was sensitive to the dispersant treatment and while its cover declined during 1986,

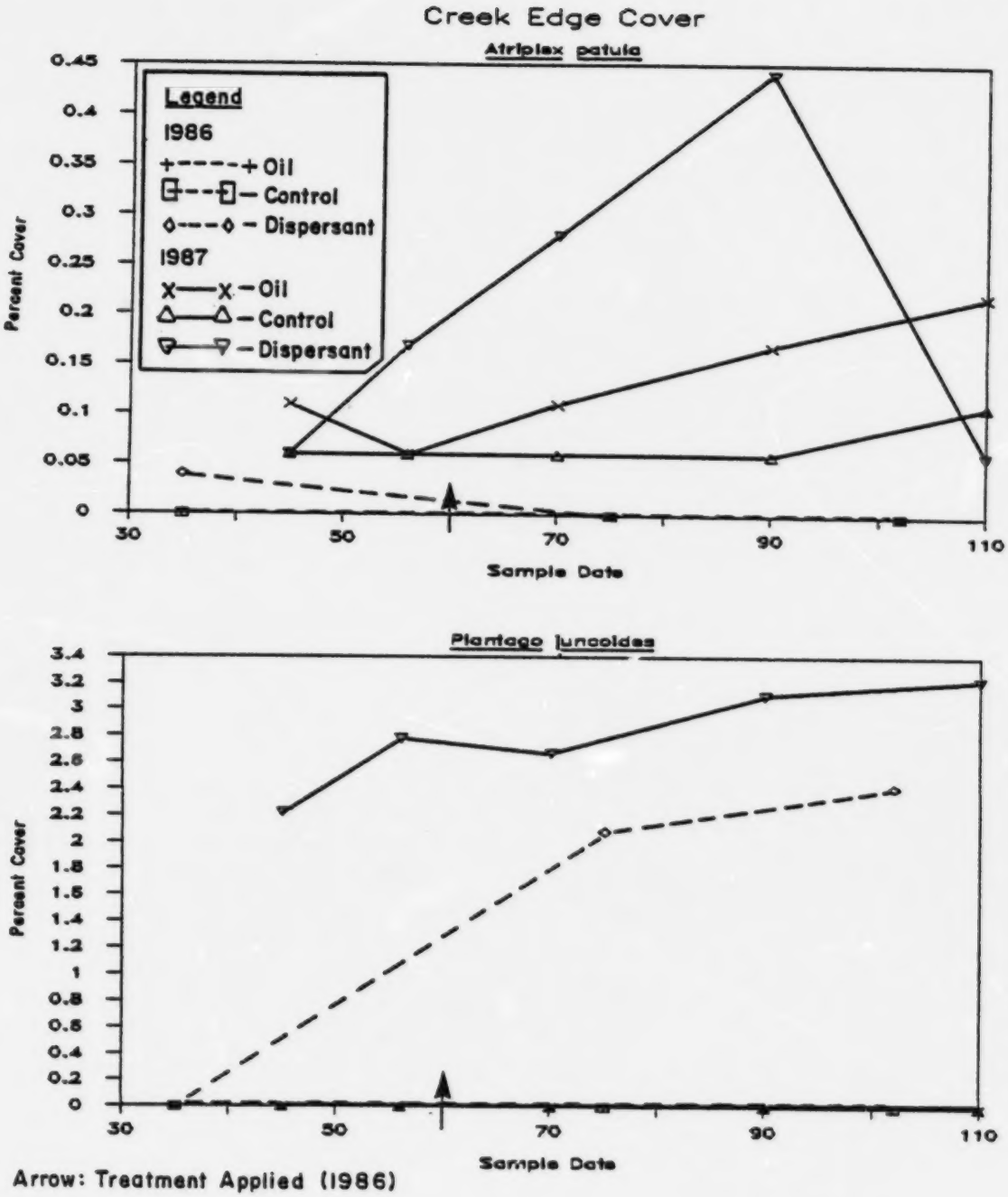


Figure 24: Plots of mean Atriplex patula and Plantago juncoides cover in the creek edge zone versus time.

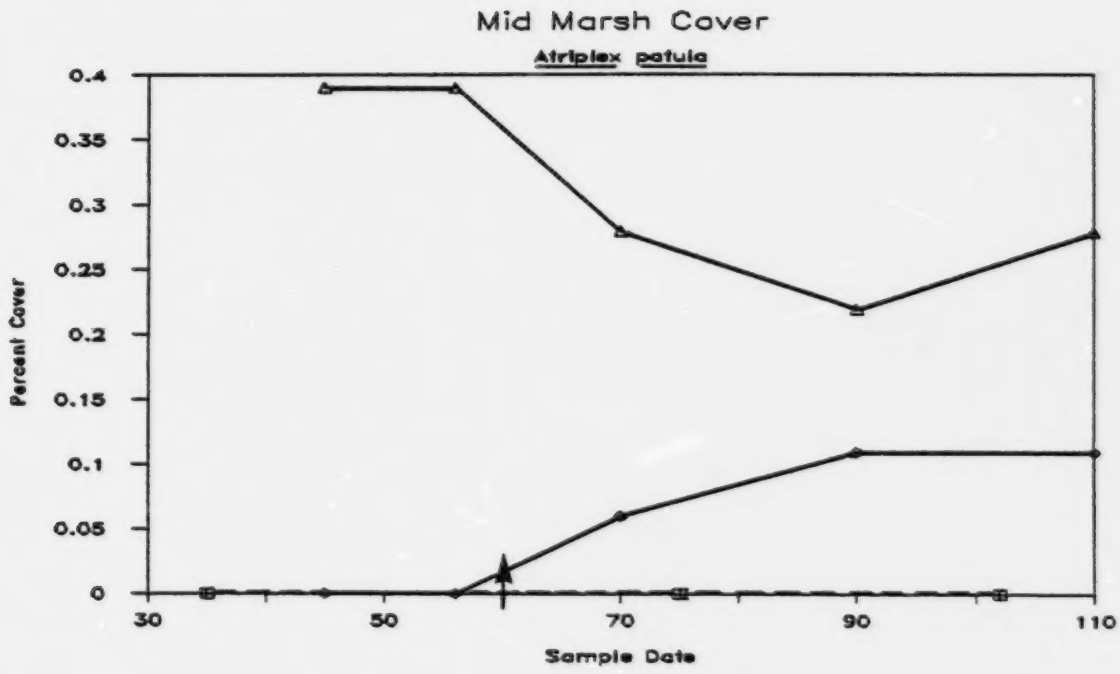


Figure 25:. Plot of mean Atriplex patula cover in the midmarsh zone versus time.

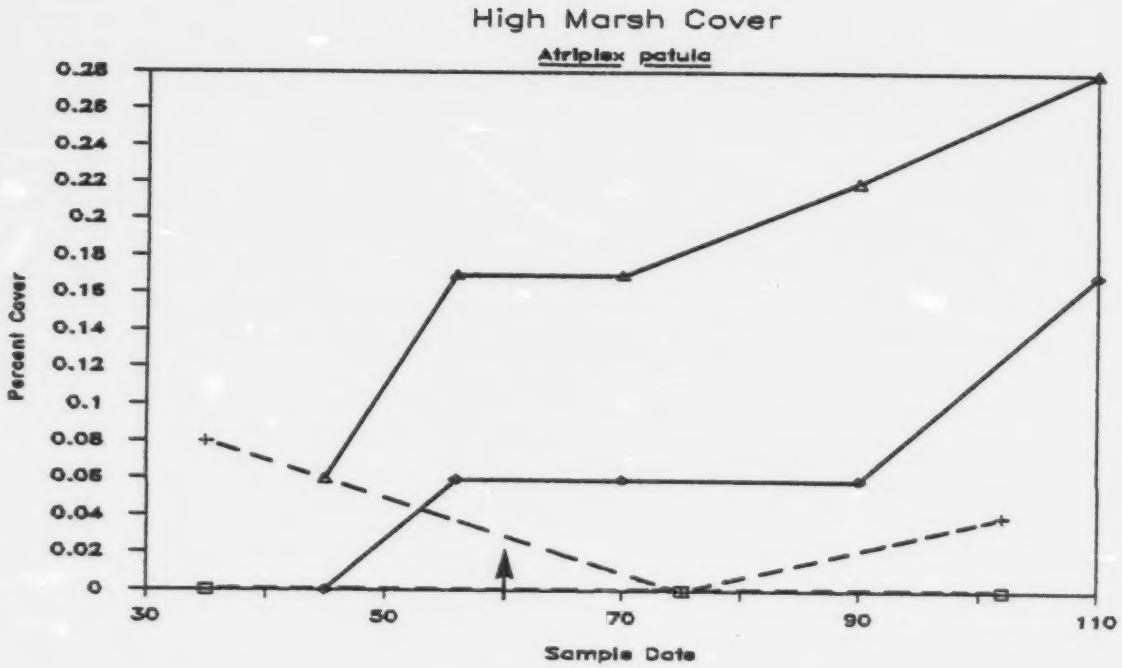


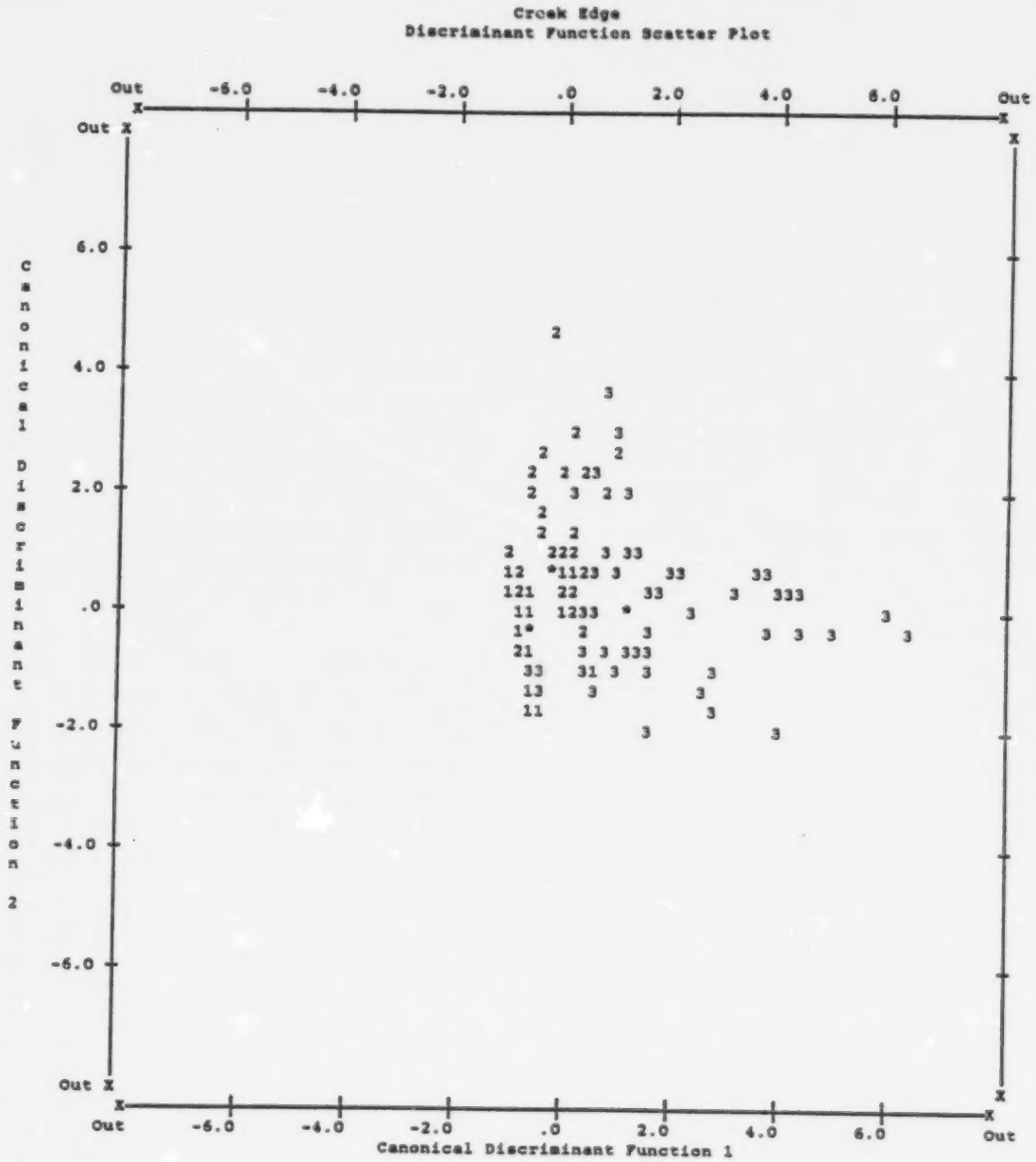
Figure 26: Plot of mean Atriplex patula cover in the high marsh zone versus time.

Plantago was able to quickly occupy the available space. Slow expansion of Plantago cover in 1987 probably reflects increased competition from the recovering S. alterniflora sward.

Discriminant analyses were conducted on the cover data collected in each vegetation zone. The analysis for the Creek edge zone included control, oil and dispersant treatments, therefore, the results were presented in two dimensions. Figure 27 presents a scatter plot for all samples with detritus excluded. The first axis separated dispersant treated plots (represented by 3's) from oiled plots (2's) and control plots (1's). The second function separated oiled plots from control and dispersant plots. Separation was highly significant along both axes (Wilks' lambda. $p<0.00005$). The first axis accounted for 72% of the variance explained, while the second accounted for 28%. Table 4 presents the coefficients of the standardized canonical discriminant function and the correlation of each species with each function. The asterisks with each correlation indicate which axis the species is most closely associated. For function 1 which separated dispersant treated plots from other treatments, Suaeda maritima, Plantago juncoides and Salicornia europaea were most important. Separation of the control and oil treated plots was most strongly determined by Spartina alterniflora and S. patens. For each point the probability of belonging to each treatment can be assessed. Each data point can be assessed as to whether it matches its treatment, or is more similar to another treatment. Counting the number that are assessed as misclassified provides an impression of the separation (or overlap) of the ranges resulting from treatment. For the creek edge zone, without standing dead cover incorporated, 97 of the 255 plots were considered misclassified.

The analysis which included standing dead yielded similar results. Both axes were highly significant with detritus associated with the second (control-oil) axis. The inclusion of standing dead cover in the analysis increased both the total variance and the proportion explained by axis 2.

These results indicate that the dispersant treatment was responsible for the largest floristic changes in the creek edge zone. These floristic changes were produced by the proliferation of annual species such as Salicornia and Suaeda after severe damage to the Spartina sward. The small degree of variance accounted for by the oil treatment (28%), indicated how little impact the oil treatment had on the creek edge zone. The results of the analysis suggest that the oil treatment had relatively little effect on species composition since the species most correlated with this function were the dominant Spartina species.



Symbols used in Plots

Symbol	Label
1	Control
2	Oil
3	Dispersant
*	Group Centroids

Figure 27: Scatter plot of discriminant analysis scores for plot visits in the creek edge zone. Species cover data was used in the analysis.

The high marsh and midmarsh zones involved only two treatments (control and oil), consequently, only one function can be generated to separate them. In the analysis for the midmarsh zone, separation was highly significant (Wilks'Lambda,$P < 0.00005$) with S. alterniflora having the only strong correlation (Table 4). Figure 28 clearly indicates the degree of separation obtained. A total of 19 out of 162 sampling units were flagged as misclassified. The results for the analysis incorporating standing dead cover were similar. Standing dead cover was negatively correlated with the function.

The high degree of separation for the midmarsh data indicates the severity of oil toxicity in this zone. High correlation with only S. alterniflora indicated both the high degree of oil induced mortality associated with this species and the relatively poor performance of Salicornia and Suaeda in this zone.

The results for the high marsh zone were also highly significant (Wilks' Lambda, $P<0.00005$), but showed more overlap (Figure 28) as indicated by the higher proportion of misclassifications (48 of 162). Salicornia europaea was highly correlated with the function, while Atriplex patula had a moderate positive correlation and Spartina alterniflora had a moderate negative correlation. Spartina patens showed little correlation with the function (Table 4). When standing dead cover was included in the analysis the number of misclassifications was reduced to 29 out of 162. The resultant function was less strongly correlated with Salicornia europea (0.65) and the correlation with standing dead was 0.44.

The high degree of misclassification for this zone suggested that the oil treatment had less impact on the floristic composition of this zone than did the oil treatment in the midmarsh zone or the dispersant treatment in the creek edge zone. High correlation of Salicornia with the function indicates that the oil treatment caused enough damage to the S. patens sward to permit a substantial increase in the abundance of Salicornia.

Fluorometry

The 1986 study (Lane et al., 1987) consisted of a field study and a parallel greenhouse study. Fluorescence induction measurements were made only for plants from the greenhouse study as a result of logistic constraints. Fluorometric analysis proved to be a sensitive technique for the detection of physiological stress in Spartina. It was decided to use this technique in the 1987 field study to determine whether plants were still stressed by the treatments one year after their application.

Table 4. Standardized canonical discriminant function coefficients (SCDFC) and pooled-within-groups correlations between species and canonical discriminant functions (CORR). Species cover values were used in the analyses for the three vegetation zones.

CREEK EDGE

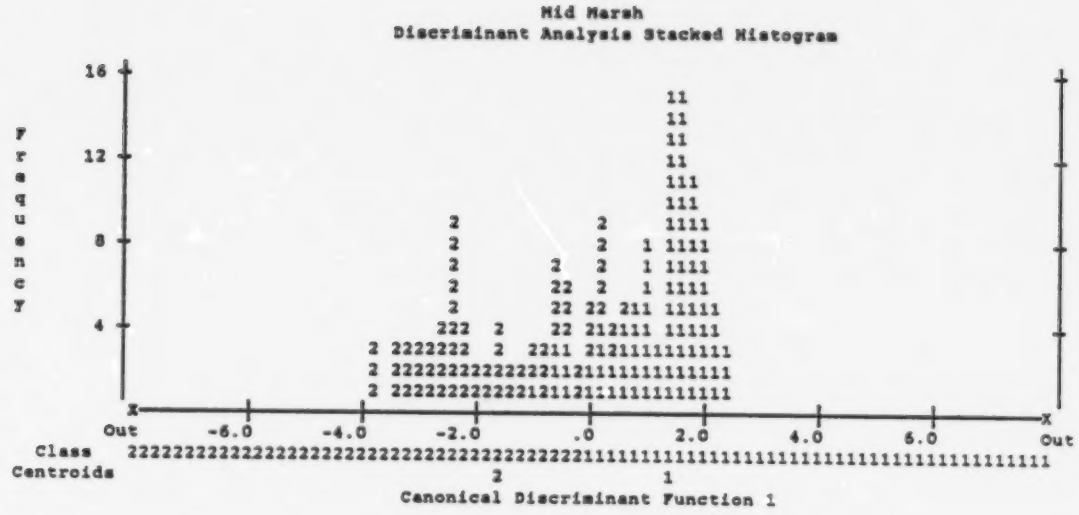
	SCDFC		CORR	
Species	Function 1	Function 2	Function 1	Function 2
Spartina alterniflora	-0.108	0.659	-0.197	0.659*
Spartina patens	0.153	0.684	0.052	0.690*
Salicornia europaea	-0.190	0.259	0.605*	0.247
Atriplex patula	-0.006	0.086	0.254	0.189
Suaeda maritima	0.918	-0.019	0.885*	0.084
Plantago juncoides	0.437	-0.114	0.637*	-0.146
Glaux maritima	-0.020	0.028	0.193	-0.044

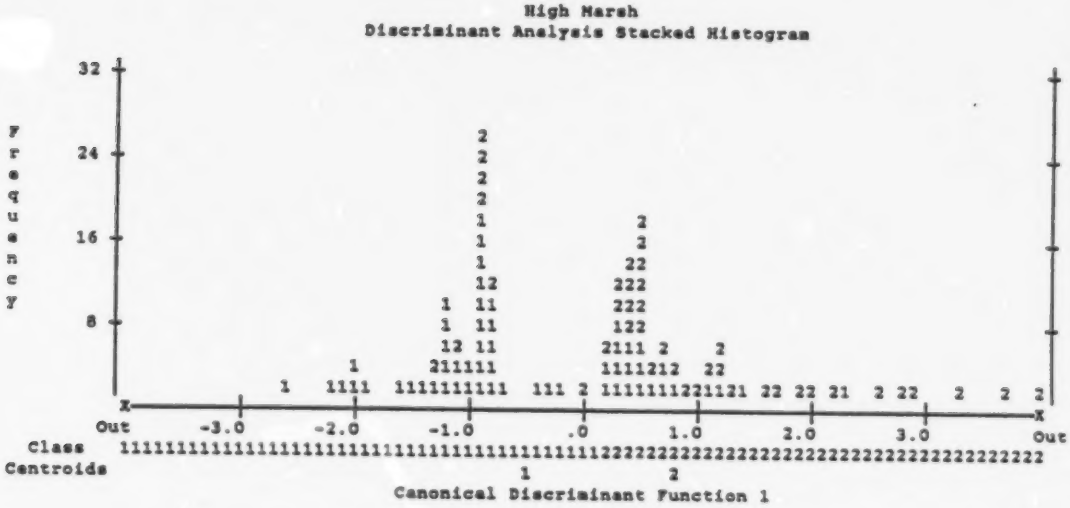
MIDMARSH

	SCDFC	CORR
Species	Function 1	Function 2
Spartina alterniflora	0.999	0.867
Spartina patens	0.432	0.115
Salicornia europaea	0.129	-0.141
Atriplex patula	-0.162	-0.287
Suaeda maritima	-0.205	-0.191
Triglochin elata	0.106	0.123
Limonium nashii	-0.061	-0.063

HIGH MARSH

	SCDFC	CORR
Species	Function 1	Function 2
Spartina alterniflora	-0.280	-0.307
Spartina patens	0.151	0.001
Salicornia europaea	0.903	0.886
Atriplex patula	0.152	0.343
Suaeda maritima	-0.109	0.102
Triglochin elata	-0.053	0.058
Festuca rubra	0.148	0.147
Glaux maritima	0.198	0.242





Symbols used in Plots

Symbol	Label
1	Control
2	Oil

Figure 28: Stacked histograms of discriminant analysis scores for plot visits in the midmarsh zones and the high marsh zone. Species cover data were used in the analyses.

Data from the greenhouse study, unfortunately, are not directly comparable to field fluormetry results from 1987 since the microcosm culture system and modifications in sampling techniques may have altered the response of the plants during the fluorescence induction testing. Comparison of the fluorometry results with the impacts of the oil and dispersant treatments on other parameters which were monitored in both the field and greenhouse experiments in 1986 (height, stem density, biomass and leaf anatomy), suggest that treatment impacts were similar in both systems. We believe, therefore, that the general trends noted for the impacts of these treatments as detected by the fluorometric techniques employed in the 1986 greenhouse study, may be indicative of the conditions which would have been encountered in the field during the 1986 growing season.

Results from the 1986 greenhouse study (Lane et al., 1987) provided evidence of damage to the photosynthetic systems of Spartina alterniflora and S. patens in all three vegetation zones. Oil treatments caused little damage in the creek edge and high marsh zones but had significant impacts in the midmarsh zone. The creek edge dispersant treatment was responsible for significant damage to Spartina alterniflora. The impacts of the oil treatment in the midmarsh zone and dispersant treatment in the creek edge zone lasted throughout the 1986 growing season.

Of the two fluorometric measures utilized in the 1986 study, the 100 second difference values (fluorescence at 100 seconds - initial fluorescence) proved to be the most sensitive measure, indicating plant stress within one day of treatment application. Peak fluorescence (Peak fluorescence - Initial fluorescence) was much less sensitive, reliably indicating plant stress only near the time of death when other symptoms (such as chlorosis) were already evident.

The fluorescence effects observed consisted of reduced peak fluorescence and a faster return to initial values. These symptoms were suggestive of a disruption of the light receptor system of the plants.

In 1987, oiled plants from the creek edge zone (Figure 29) tended to have reduced peak variable fluorescence on all sampling dates with values for oiled plants ranging between 89 and 97% of control values. None of these reductions were significant on any of the sampling dates. Peak variable fluorescence in dispersant treated plots was low on day 50 but increased to control levels by day 91. No significant differences were noted on any sampling date. One hundred second difference values for oiled plants were significantly lower than controls on day 71 ($p=0.041$, ANOVA), but were not significantly different on any other sampling date. One hundred second difference values for dispersant treated plants were significantly higher

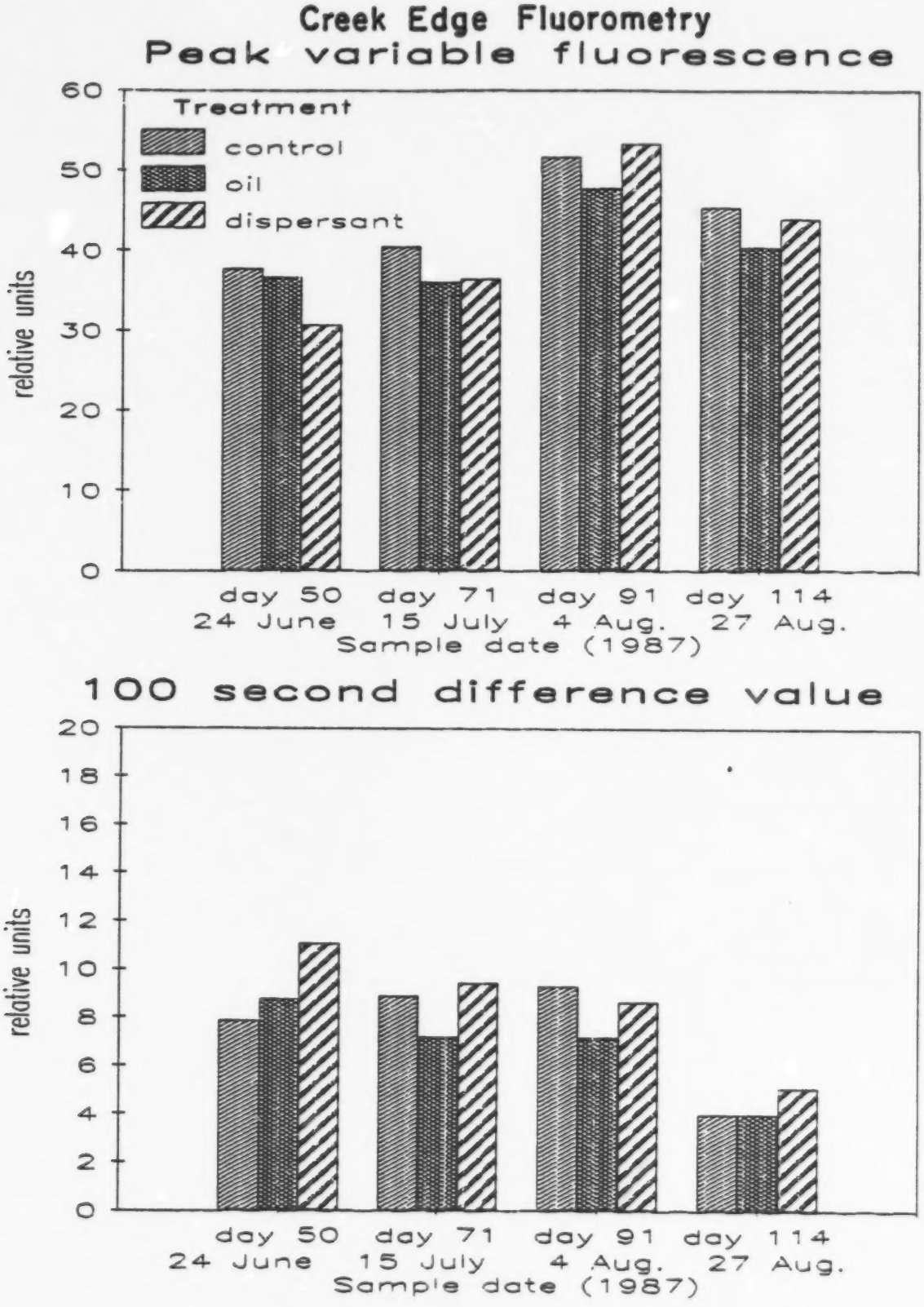


Figure 29: Histograms or peak variable fluorescence and 100 second difference values for plants from the creek edge zone on four sampling dates in 1987.

than for control plants on day 50 (p=0.015, ANOVA), however, values for all other samping dates were not significantly different from the controls. These data suggest that there was little if any residual toxicity associated with the treated plots in the creek edge zone one year after treatment applications.

In the midmarsh zone (Figure 30) oiled plants tended to have reduced peak variable fluorescence values as was noted for the creek edge zone. These reductions, however, tended to be larger than in the creek edge zone with values for oiled plants ranging between 67 and 87% of control values. Peak variable fluorescence values of oiled plants were significantly lower than values for control plants on days 71 (p=0.021, ANOVA) and 114 (p=0.004, ANOVA). The trend for the 100 second difference values was also similar to that noted in the creek edge zone. Oiled plot values were lower than control plot values on days 71 and 91 but were higher than the controls on days 50 and 114. No significant differences in 100 second difference values were noted on any of the sampling dates. The significant reductions in peak variable fluorescence indicate that residual oil may have produced some plant stress.

In the high marsh zone (Figure 31) peak variable fluorescence of oiled plants was lower than the controls on the last two sampling dates, however, no significant differences were noted on any sampling date (Appendix 8). One hundred second difference values for high marsh plants displayed a trend which was common to all vegetation zones. Oiled plants had higher values than controls on day 50 but returned to control levels on all other sampling dates. The increase on day 50 was significant (p=0.031, ANOVA). Values on all other sampling dates were not significantly different from the controls. The data suggest that there was little or no residual oil toxicity in the high marsh plots one year after treatment applications.

Fluorometric analysis suggested that oil applied to the creek edge and high marsh areas had weathered sufficiently by 1987 to eliminate oil as a source of plant stress. Similarly, the dispersant applied to the creek edge zone in 1986 caused no plant stress in 1987. The dispersant was probably removed from the study plots by tidal inundation within a month of application. Data for the midmarsh zone suggested that residual oil may have stressed the plants in that zone. Sediment oil data for 1987 were too variable to determine whether more oil was present in the midmarsh zone than in the other zones. Gas chromatography data suggested that oil in all of the zones was extensively weathered by 1987. Long term toxicity in the oiled midmarsh plots may have been attributable to the stressful nature of the midmarsh environment. Physiological stresses associated with the midmarsh, such as reduced nitrogen uptake, high root respiration and

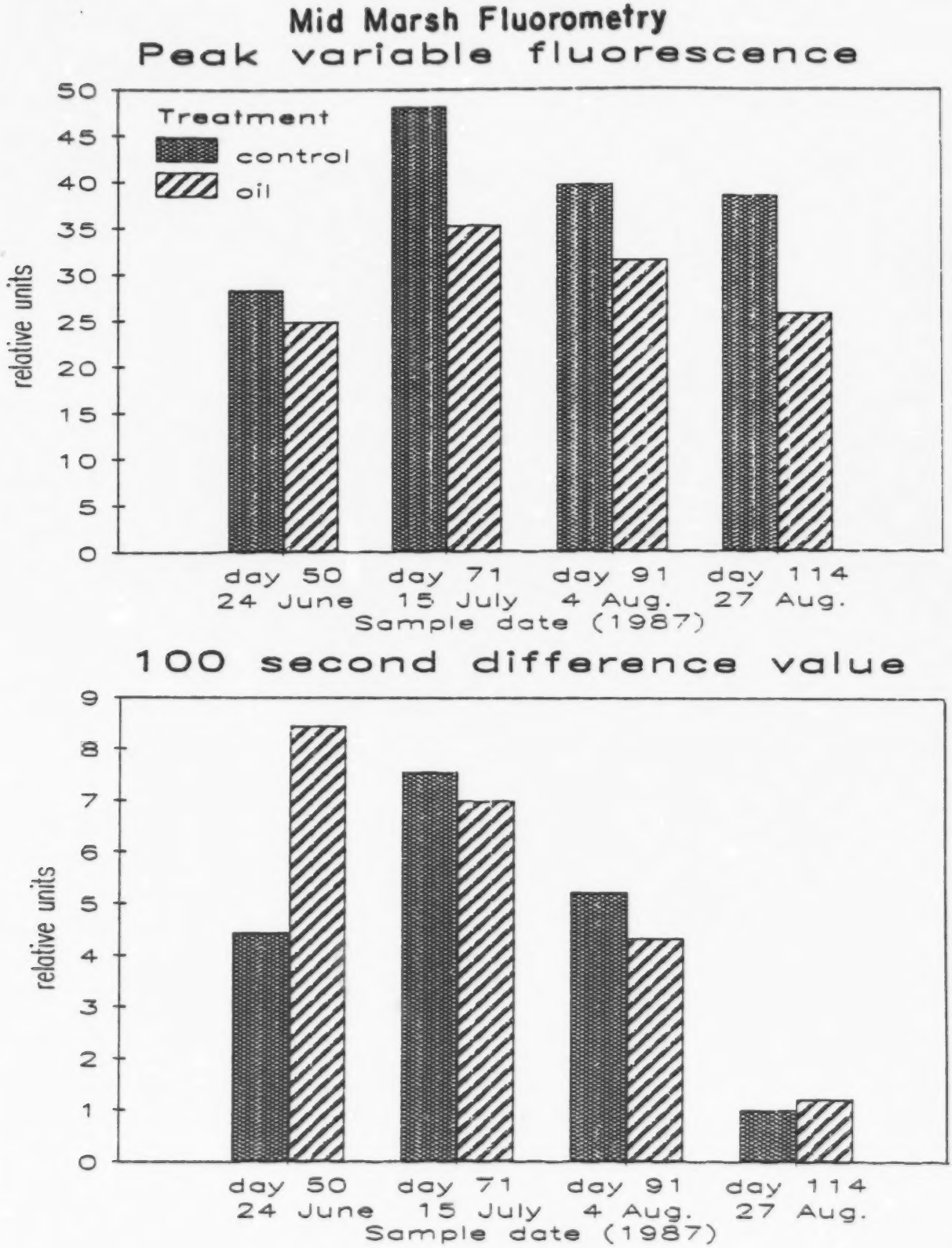


Figure 30: Histograms of peak variable fluorescence and 100 second difference values for plants from the midmarsh zone on four sampling dates in 1987.

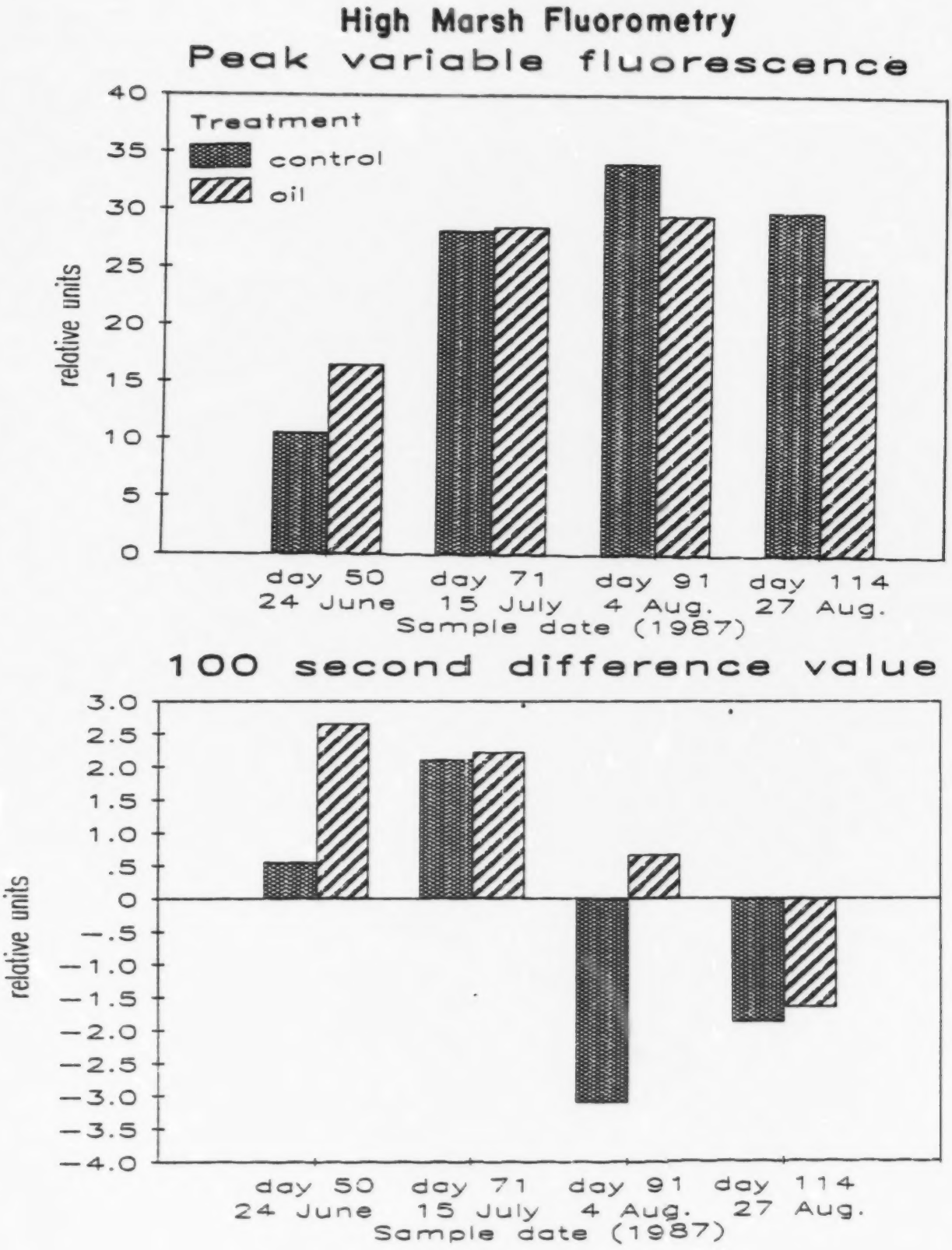


Figure 31: Histograms of peak variable fluorescence and 100 second difference values for plants from the high marsh zone for sampling dates in 1987.

hydrogen sulfide toxicity may have reduced the tolerance of midmarsh plants to oil residues which had little or no effect on creek edge or high marsh plants.

Peak variable fluorescence was the only fluorometric parameter which was consistently altered by the oil treatments. Reduction of peak variable fluorescence would suggest damage to the light receptor system of the midmarsh plants. Lower concentrations of photosynthetic pigments would reduce the proportion of the incoming light which was absorbed by these pigments and thus available for fluorescence. Chlorosis might be expected under these conditions, however, no differences in plant coloration were noted during the field observatons nor were noticeable reductions in chloroplast density observed in the leaf anatomy study. The fluorometric technique may have been sensitive enough to indicate stress before symptoms became visible or the mechanism of light receptor damage may not have involved the physical destruction of chloroplasts.

Leaf Anatomy

Anatomical data from 1986 (Lane et al., 1987), demonstrated that microscopic symptoms were identical for both oil and dispersant treated plants. Microscopic symptoms consisted of a gradual reduction in chloroplast density in conjunction with progressive chlorosis at the macroscopic level. The data suggested that both treatments had probably caused disruption of cell membranes leading to the destruction of chloroplasts and the eventual death of the cells. Additional macroscopic symptoms included oil induced stem and leaf deformities of Spartina patens which affected approximately 10% of oiled plants.

Results from the 1987 study revealed no microscopic or macroscopic differences between the control and oil or dispersant treated plants. These data suggest that the constituents of the oil responsible for the symptoms observed in 1986 had been weathered or consumed by microbes before the 1987 growing season. Any residual toxicity might be expected to produce more subtle changes in plant morphology such as reductions in growth rate or alterations in reproduction. These have been dealt with elsewhere in the text.

CONCLUSIONS

The oil treatment appeared to have been responsible for a reduction in the thickness of anaerobic reducing zones in all three vegetation zones. Significant reductions were noted only in the high marsh zone. Declines in reducing zone thickness may have been caused by oil induced decreases in heterotrophic activity which permitted the downward migration of the aerobic zone.

Hydrocarbon concentrations in the salt marsh sediments were highly variable, however, temporal trends in concentrations were apparent. Hydrocarbon concentrations in all zones initially increased over a period of approximately 30 days as oil was removed from vegetation and deposited on the sediment. In the creek edge and high marsh zones, concentrations peaked then declined within 1986 as a result of weathering and possibly flushing of oil from the sediments. Concentrations in the midmarsh zone remained high throughout 1986. In 1987, concentrations in the creek edge and high marsh zones declined relatively little and were similar to or slightly lower than end of growing season concentrations in 1986. Hydrocarbon concentrations in the midmarsh zone declined substantially during the 1987 growing season. Most hydrocarbons under C-10 had been lost from the sediments by the end of 1986. At the end of the 1987 growing season, hydrocarbons under C-14 were absent from the sediments. Oil penetration was mostly restricted to the upper 5 cm of the sediment although slightly elevated concentrations were found between 5 and 10 cm.

Plant height data indicated that Spartina height growth in the creek edge zone was relatively unaffected by the oil treatments in both 1986 and 1987 while height growth in the midmarsh and high marsh zones were significantly reduced in 1986. The midmarsh zone was more severely affected by the oil treatments than the high marsh zone. Neither zone appeared to recover during the second growing season. Height growth in the dispersant treated plots in the creek edge zone was significantly reduced during the 1986 growing season but recovered to control levels by the 1987 growing season.

Oil impacts on stem density were still evident in the midmarsh and highmarsh zones in 1987. The largest impacts were associated with the midmarsh zone in both 1986 and 1987. The larger reductions in midmarsh stem density as compared to the creek edge zone were probably attributable to the naturally higher levels of plant stress associated with the midmarsh zone. The additional stress of the oil spill probably taxed the metabolism of the midmarsh plants to the point where most individual stems died. It was difficult to determine the degree of recovery in the creek edge zone

since increases in stem density of oiled plots in 1987 were accompanied by large reductions in control stem density. Nevertheless, it appeared that recovery of stem density was most advanced in this zone. The dispersant treatment caused a large reduction of stem density in the creek edge zone in 1986. A moderate increase in stem density was noted for this treatment in 1987.

The density of flowering shoots was substantially reduced (although not significantly) in 1986 by both the oil and dispersant treatments in all zones. In 1987, flowering shoot densities were high in the treated plots, particularly those in which heavy mortality in 1986 was followed by substantial vegetative recovery in 1987. This response was probably the result of a reduction of intraspecific competion which increased the proportion of resources available for flowering. Flowering shoot densities in the creek edge and midmarsh control plots declined in 1987, probably in response to drought conditions experienced during the 1987 growing season.

Total above-ground living biomass data for 1986 indicated that the midmarsh zone was severely affected by the oil treatments while the creek edge zone was virtually unaffected. Total above-ground living biomass in the high marsh zone was moderately reduced. Data for 1987 revealed full recovery for the creek edge zone. Total above-ground living biomass in the midmarsh zone increased significantly while recovery in the high marsh zone was rather poor. Total above-ground living biomass in the dispersant treated creek edge plots was significantly reduced in 1986 but returned to control levels by the end of the 1987 growing season.

Standing dead biomass data collected in 1986 generally reflected the degree of mortality associated with the various treatments and zones; however, it appeared that standing dead biomass values in the oiled creek edge and midmarsh zones were larger than expected from the observed stem density reductions. These data suggested that decomposition had been retarded by the oil treatments. The data revealed that the high marsh zone naturally maintained a very high standing crop of dead stems (up to 2 times higher than total above-ground living biomass). Standing dead biomass returned to expected values in all oiled plots in 1987. In the creek edge dispersant plots in 1987 standing dead biomass was lower than expected as a result of unusually low stem mortality during the 1987 growing season.

Two annual species, Salicornia europaea and Suaeda maritima, increased their biomass in response to mortality of the dominant Spartina species in all three vegetation zones. These increases did not occur until the 1987 growing season either as a result of a lack of seed following the period of high Spartina mortality or inhibition of Salicornia and Suaeda seed

germination and/or seedling development by the oil during the 1986 growing season.

Flower biomass was reduced by both the oil and dispersant treatments in 1986, however, in 1987 flower biomass in the treated plots increased greatly with the largest increases associated with the creek edge dispersant plots where mortality had been high in 1986 but recovery had been rapid in 1987. This response was probably attributable to increased availability of resources caused by decreased intraspecific competition in plots where stem density reductions had occurred in 1986. Flower biomass in the creek edge and midmarsh control plots declined in 1987 possibly as a result of drought conditions.

Mean weight of individual Spartina shoots was not affected by the oil treatments in the creek edge zone in 1986, however, it increased relative to control plants in the midmarsh and high marsh zones as a result of selective mortality of small plants. Dispersant treated plants in the creek edge zone in 1986 weighed only half as much as control plants. In 1987, they were heavier than the control plants probably as a result of reduced intraspecific competition. The mean weights of oiled creek edge and high marsh plants were similar to their respective controls in 1987 indicating little residual toxicity. Midmarsh plants were twice as heavy as control plants in 1987 but were significantly shorter. It is believed that morphological changes may have been induced by the oil treatment.

Cover data collected during the 1986 growing season indicated little oil induced mortality in the creek edge zone, moderate mortality in the high marsh zone and severe mortality in the midmarsh zone. The dispersant treatment caused moderate mortality in the creek edge zone. Data from 1987 indicated little expansion of the Spartina sward in any of the zones. It was difficult to determine the degree of recovery in the creek edge zone since there was a considerable reduction in the cover of control plots between 1986 and 1987. It was impossible to determine whether the factor or factors responsible for the decline of the control cover would also act upon the treated plots. Drought conditions experienced in 1987 may have been responsible for the decline in control cover. Stress induced by the treatments may have been reduced during the 1987 growing season but was replaced by drought stress resulting in the lack of recovery noted in this zone.

Annual halophytes such as Salicornia europaea, Suaeda maritima and Atriplex patula were rather rare in 1986. Oil and dispersant treatments generally resulted in reductions in their abundance during 1986. In 1987, large increases in the cover of these species were noted in the oil and

dispersant treated plots. High negative correlation coefficients between the cover of the dominant Spartina species in each zone and the cover of these annual species suggested that these species were rapidly colonizing areas where Spartina had been eliminated. Colonization was rapid in the creek edge zone but was relatively slow in the midmarsh and high marsh zones. In the midmarsh zone poor soil aeration and the physiological problems associated with it may have reduced the rate of colonization while in the high marsh zone the presence of a large standing crop of standing dead shoots may have reduced the rate of colonization of annuals through the shading of shade intolerant seedlings.

Fluorometry data indicated that plant stress levels were similar for control and oil or dispersant treated plots in the creek edge and high marsh zones, suggesting that there was no residual toxicity associated with these treatments in 1987. In the midmarsh zone there appeared to be evidence of some residual oil toxicity in 1987. The fluorescence response of these plants was indicative of damage to the light receptor system.

Leaf anatomy results from 1986 revealed a progressive reduction in the density of chloroplasts in the outer bundle sheath and mesophyll cells in both oil and dispersant treated Spartina alterniflora and S. patens which was accompanied by the development of chlorosis at the macroscopic level. Leaf and stem deformities were also noted for Spartina patens. In 1987, no microscopic or macroscopic differences were found between control and treated plants. These results suggest that there was little residual toxicity associated with the oil or dispersant treated plots in 1987.

This study has demonstrated that sensitivity to crude oil varies considerably among the various plant communities in the salt marsh. The creek edge zone, dominated by tall Spartina alterniflora was quite tolerant of the crude oil treatment. Results from 1986 reveal only small reductions in height, stem density, biomass and cover and by 1987 no evidence of toxic effects were apparant. Similar results have been found in several other studies (Delaune et al, 1979; Webb et al., 1981; Ferrel et al., 1984; and Webb and Alexander, 1985). The short Spartina alterniflora of the mid marsh zones, however, was very susceptable to the oil treatment. In 1986 all parameters investigated were negatively affected by the oil. In 1987 only limited recovery of this zone was noted. The stressful nature of the midmarsh zone predispose it to severe impacts from additional perturbations such as oil toxicity. In addition, the poor drainage of the zone reduces the rate at which the oil is flushed from the sediments or metabolized by microbes further contributing to the persistence of toxic effects in this zone. The impacts reported for the midmarsh zone illustrate the degree to which existing environmental stresses can affect the ability of a species

or community to cope with pollutants.

The high marsh zone was intermediate in its response to the oil treatments. Most parameters demonstrated significant impacts in 1986, however, these impacts were not as catastrophic as observed in the midmarsh zone. Relatively little recovery was noted in the various parameters in 1987. This lack of recovery may be at least partially the result of the slow vegetative colonization of open patches and poor seed set which are normally associated with this species.

Exposure of the creek edge zone to the disperant resulted in considerably more damage than the crude oil. The toxic effects, however, were short lived and most parameters investigated in this study had returned to normal or were rapidly recovering one year after the dispersant was applied.

All three vegetation zones demonstrated similar successional trends after exposure to crude oil . Annual species of Chenopodiaceae invaded the treated plots two years after oiling. Salicornia europaea and Suaeda maritima were the most important species. Their abundance was inversely correlated with Spartina cover. We anticipate that the cover of these species will probably increase for two years following treatment, then decline as Spartina develops a dense sward. Based on current rates of recovery, the creek edge zone recovered from oiling within one year of treatment and will probably recover from the dispersant treatment within two years. The high marsh zone will probably require three to four years for full recovery, while the mid marsh zone will probably require at least five years.

The results suggest that lightly oiled marshes can recover relatively quickly. Natural cleaning may be ecologically preferred if oil contamination is low and/or when the oil enters only the creek edge zone. The use of relatively non-destructive clean up techniques such as low pressure flushing of the marsh surfaces may be justifiable if substantial portions of the midmarsh zone were oiled, provided trampling damage associated with the clean up technique was minimal. Oil induced damage to the high marsh zone, although significant, probably does not warrant the implementation of clean up techniques. The productivity of this zone was significantly reduced by exposure to the crude oil, however, the community structure of this zone remained intact and productivity can probably be expected to return to normal in a few years.

APPENDIX I: PRINCIPLES OF FLUORESCENCE INDUCTION MEASUREMENTS

Light absorbed by the chloropyll-a reaction center of photosystem II (the oxygen evolving part of the photosynthetic system) excites ground state electrons in the chlorophyll molecule. An excited electron very quickly (within ca 10^{-10} milliseconds) loses some of its excitation energy, which is dissipated as heat. This less excited, lower energy state may then jump to a primary electron acceptor, or it may return to its ground state. When it does the latter, its energy is dissipated as fluorescent light reemitted from the molecule, or as heat. If the electron is passed to the primary electron acceptor, the lost electron is replaced by an electron taken from water (with concommittant release of free oxygen). The primary electron acceptor in the meantime loses the electron to an electron carrier which loses it to another carrier and so on through photosystem I, until finally an electron is passed to NADP. The electron carrying-NADP then moves out of the internal membranes of the choroplast where the chlorophyll and carriers are located, into the stroma where it participates in the reactions which convert carbon dioxide to sugar.

Because some of the excited electron energy is always lost as heat, and because there is an inverse relationship between the wavelength of light and its energy, the wavelength of light reemitted as fluorescence is longer than that of the light that was absorbed. The fluorescence wavelength is characteristic of the molecule; for chlorophyll-a under physiological conditions, there is a main band maximum at 685 nm. To measure fluorescence, the sample is subjected to light of lower light intensities, and light emission at 685 nm is monitored.

What is termed "variable fluorescence" can be thought of as an overflow phenomenon; when there is more light absorbed than can be immediately processed via the chemical route, part or most of the excited electron energy is dissipated as fluorescence.

When the plant is first exposed to light, the initial electron acceptor and the downstream carriers are empty handed and can readily receive electrons from chlorophyll. As they become saturated with electrons, their ability to process more electrons decreases, the chloropnyll reaction center cannot pass on its excited electrons, the excited electrons drop back to their unexcited ground state and fluorescence rises. It reaches a peak value within about 3 seconds of the light being turned on. During this period, the rest of the electron carrier system and the Calvin cycle become activated and begin to accept electrons from the upstream carriers and the primary electron acceptor. More electrons can be processed, therefore,

fluorescence begins to decline. Within about 10 seconds, a steady state in the processing system is established, and there is a relatively low, steady level of fluorescence (Figure 2).

The pattern of fluorescence change to this point is called the fluorescence induction curve. Over longer periods (minutes), there may be another, slower increase in fluorescence and a new plateau in fluorescence reached as processes further downstream from the initial events become saturated and begin to regulate the upstream flow.

Fluorescence induction is a sensitive probe for photosynthesis over very short periods, and responds to any changes affecting photosystem II activity. Thus, it has been very important in elucidation of photosynthetic mechanisms (Papageorgiou, 1975). More recently, it has been used in studies investigating the effects of various types of stress on plants. Many types of stress - water, heat, excess light, salt, chilling, freezing - result in changes in fluorescence induction characteristics long before there are changes in other measureable characteristics (Smillie & Hetherington, 1983). There appear to be no studies on stress to foliage that have indicated effects on other processes without there being effects on fluorescence induction.

Changes in fluorescence curves brought about by stress can be interpreted to provide presumptive evidence about the way in which the stress is affecting photosynthesis. For example, a reduced peak in fluorescence could indicate damage to the light receptor system; increased peak fluorescence could be due to damage on the carrier side of PS II close to the reaction centre, and a slow decline from the peak could be due to damage further downstream.

To our knowledge, this technique has not been previously used in studies of oil and/or dispersant damage to plants. We had several objectives in applying it in this study:

i) We wished to determine if any of the treatments affected the photosynthetic system, and how such effects varied between treatments and zones.

ii) By comparing the fluorescence induction parameters with other criteria of stress (growth rates, flowering, etc.) measured at different times, we wished to determine if this method could be used to forecast stress effects that would otherwise be apparent only after longer intervals.

iii) Assuming that changes in the chlorophyll induction curve would be one of the first responses to stress, we wished to use the method to indicate how quickly after applying oil and/or dispersant, the plants begin to be stressed.

APPENDIX 2: SUMMARY TABLE OF ANALYSIS OF VARIANCE RESULTS FOR THE THICKNESS OF REDUCING ZONES

One way analysis of variance probabilities. Data were untransformed. Oil was applied on day 60 in 1986.

	Creek Edge	Midmarsh	High Marsh
1987			
Day 45	0.284	0.162	0.000
Day 75	0.568	0.172	0.627
Day 110	0.162	0.786	0.571

APPENDIX 3: SUMMARY TABLE OF ANALYSIS OF VARIANCE RESULTS FOR PLANT HEIGHT DATA

One-way analysis of variance probabilities. Data were transformed by ln((x/14)+1). Treatments were applied on day 60 in 1986.

	Creek Edge		Midmarsh	High Marsh
	Oil	Dispersant	Oil	Oil
1986				
Day 26	0.000	0.404	0.000	0.006
Day 34	0.001	0.000	0.128	0.425
Day 49	0.000	0.000	0.133	0.455
Day 57	0.041	0.692	0.245	0.000
Day 75	0.601	0.000	0.400	0.000
Day 89	0.043	0.000	0.020	0.000
Day 102	0.179	0.000	0.199	0.055
Day 123	0.034	0.000	0.024	0.000
1987				
Day 26	0.000	0.000	0.095	0.000
Day 48	0.000	0.021	0.001	0.486
Day 67	0.065	0.126	0.000	0.000
Day 90	0.351	0.785	0.000	0.000
Day 115	0.034	0.455	0.000	0.000

APPENDIX 4: SUMMARY TABLE OF ANALYSIS OF VARIANCE RESULTS FOR STEM DENSITY DATA

One-way analysis of variance probabilities. Data were transformed by ln(x +1). Treatments were applied on day 60 in 1986.

	Creek Edge		Midmarsh	High Marsh
	Oil	Dispersant	Oil	Oil
1986				
Day 26	0.875	0.000	0.625	0.046
Day 49	0.190	0.000	0.039	0.172
Day 89	0.000	0.000	0.000	0.000
Day 123	0.043	0.000	0.000	0.000
1987				
Day 26	0.477	0.000	0.000	0.848
Day 48	0.187	0.106	0.000	0.052
Day 67	0.168	0.000	0.000	0.021
Day 90	0.926	0.162	0.000	0.073
Day 115	0.014	0.031	0.000	0.019

APPENDIX 5: SUMMARY TABLE OF ANALYSIS OF VARIANCE RESULTS FOR STEM DENSITY OF FLOWERING SPARTINA PLANTS

One-way analysis of variance probabilities. Data were transformed by ln(x + 1). Treatments were applied on day 60 in 1986.

	Creek Edge		Midmarsh	High Marsh
	Oil	Dispersant	Oil	Oil
1986	0.353	0.357	0.274	0.138
1987	0.134	0.004	0.130	0.525

APPENDIX 6: SUMMARY TABLE OF ANALYSIS OF VARIANCE RESULTS FOR THE BIOMASS DATA

One-way analysis of variance probabilities. Data were transformed by square root. Treatments were applied on day 60 in 1986.

	Creek Edge				Midmarsh		High Marsh	
	Oil 1986	Oil 1987	Disp 1986	Disp 1987	Oil 1986	Oil 1987	Oil 1987	Oil 1986
Standing Dead Biomass	0.022	0.400	0.397	0.054	0.009	0.144	0.841	0.184
Spartina alterniflora biomass	0.613	0.488	0.017	0.685	0.003	0.096	-	0.374
Spartina patens biomass	-	0.374	-	-	0.374	0.271	0.221	0.010
Salicornia europaea biomass	-	0.786	0.374	0.370	-	0.605	-	0.978
Suadea maritima biomass	-	0.774	-	0.581	-	0.997	-	-
Atriplex patula biomass	-	0.374	-	0.374	-	-	-	-
Total Above-ground Living Biomass	0.613	0.480	0.017	0.627	0.003	0.115	0.221	0.011
Mean Weight Per Shoot	0.980	0.820	0.421	0.389	0.624	0.099	0.244	0.799
Total Biomass of Flowers	0.322	0.193	0.378	0.006	0.148	0.136	0.225	0.376

APPENDIX 7: SUMMARY TABLES OF ANALYSES OF VARIANCE RESULTS FOR PLANT COVER DATA

One way analysis of variance probabilities. Data were transformmed by square root. Treatments were applied on day 60 in 1986.

	Creek Edge		Midmarsh	High Marsh
	Oil	Dispersant	Oil	Oil
1986				
Spartina alterniflora				
Day 35	0.139	0.028	0.609	0.328
Day 75	0.002	0.000	0.000	0.328
Day 102	0.000	0.000	0.000	0.328
Spartina patens				
Day 35	-	-	0.177	0.048
Day 75	-	-	0.328	0.304
Day 102	-	-	0.634	0.005
Salicornia europaea				
Day 35	0.200	0.666	0.430	0.014
Day 75	0.094	0.595	0.193	0.137
Day 102	1.000	0.007	0.053	0.001
Suaeda maritima				
Day 35	1.000	0.229	-	0.028
Day 75	-	-	-	0.328
Day 102	-	-	0.328	-

	Creek Edge		Midmarsh	High Marsh
	Oil	Dispersant	Oil	Oil
1987				
Spartina alterniflora				
Day 45	0.246	0.003	0.000	0.332
Day 56	0.004	0.000	0.000	0.332
Day 70	0.052	0.219	0.000	0.332
Day 90	0.150	0.395	0.000	0.332
Day 110	0.592	0.000	0.000	0.332
Spartina patens				
Day 45	-	-	0.167	0.011
Day 56	-	-	0.400	0.115
Day 70	-	-	0.346	0.001
Day 90	-	-	0.315	0.063
Day 110	-	-	0.157	0.031
Salicornia europaea				
Day 45	0.853	0.876	0.570	0.001
Day 56	0.672	0.346	0.788	0.278
Day 70	0.044	0.000	0.116	0.001
Day 90	0.023	0.000	0.146	0.000
Day 110	0.069	0.000	0.202	0.000
Suaeda maritima				
Day 45	0.276	0.002	-	1.000
Day 56	0.030	0.000	1.000	0.555
Day 70	0.188	0.000	0.044	1.000
Day 90	0.291	0.000	0.125	0.555
Day 110	0.310	0.000	0.011	0.470

APPENDIX 8: SUMMARY TABLE OF ANALYSIS OF VARIANCE RESULTS FOR PLANT VARIABLE FLUOROMETRY DATA

One-way analysis of variance probabilities. Data were transformed by square root. Treatments were applied on day 60 in 1986.

	Creek Edge		Midmarsh	High Marsh
	Oil	Dispersant	Oil	Oil
100 Second Difference Values				
Day 47	0.474	0.015	0.212	0.031
Day 68	0.041	0.522	0.534	0.927
Day 88	0.109	0.617	0.327	0.895
Day 111	0.636	0.232	0.675	0.879
Peak Variable Fluorescence				
Day 47	0.426	0.074	0.290	0.085
Day 68	0.465	0.338	0.021	0.900
Day 88	0.182	0.676	0.131	0.513
Day 111	0.304	0.830	0.004	0.263

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